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THE TRACE ELEMENT UPTAKE AND CONTENT OF PASTURE CROPS AS INFLUENCED BY EXTERNAL FACTORS

De invloed van uitwendige faktoren op de opname en het gehalte van sporenelementen in grasland

door

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الى من اهديانى قلبا هما .... نورا يضى° لى طريقى
الى من اهديانى مبادئهما .... مثلا اعلى للفضيله والصبر والتضحيه

الى من اعطا دائما .... ولم يأخذا

الى ابويّ

أُهدى هذة الرساله عرفانا بالجميل ورمزا لأجلّ ايات المحبه والتقدير .

When you reach the end of what you should know, you will be at the beginning of what you should sense

KAHLIL GIBRAN

(Sand & Foams 1929)

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CONTENTS

INTRODUCTION

Striving to overcome nutrient deficiencies in order that yields may more nearly approach the genetic limit of crop plants, (SAMUEL et al. 1966), requires an adequate supply of plant nutrients.

A large number of elements are involved in the growth and reproduction of plants and animals. Of these nutrients only a few are required in large amounts. Deficiencies of the remaining ones, which are necessary in trace amounts are most frequently related to specialized crops on certain types of soil, (HODGSON, 1963). Cropping systems become more intensive, changes in soil management practices frequently alter micronutrient availability and depletion of nutrients not added in fertilizers becomes more frequent. Once the demand for higher yield increases through major elements is more efficiently met, other nutrients are more likely to become limiting. With emphasis on high yields, an awareness of possible concomitant changes in crop quality is needed. From these various considerations one might well anticipate a renewel of interest in the role micronutrient elements may play in restricting maximum production and optimum quality of farm crops, (HODGSON, 1963).

The complicated mechanism of the nutrient uptake and metabolism has not yet enabled the assessment of suitable techniques for the study of causal relationships. The elucidation of the functions of micronutrients in plant growth therefore is wide field for research.

The approach made in this thesis indicates the importance of the trace element composition as influenced by certain external factors such as major element application in long and short term experiments, trace elements supplementation, organic and inorganic fertilization, and finally the problem of pollution in

grassland.
Chapter 1 is giving the literature review concerning the present study.
Chapters 2, 3 and 4 are relating experiments on interactions between the major elements, trace elements, organic and inorganic fertilizations and trace elements uptake by herbage crops.
Chapter 5 adds more information on the relationship between major and trace elements on long and short term field experiments, when other factors are also involved.
Chapter 6, contributes to the knowledge of the important problem of contamination in Belgian grassland.

CHAPTER I

TRACE ELEMENTS IN RELATION TO SOIL, PLANT AND ANIMAL NEED

1. INTRODUCTION :

The trace elements regarded as essential in plantnutrition are : Iron; Manganese ; Zinc; Copper ; Boron ; Molybdenum and Chlorine.

Numerous factors influence the availability of trace-elements in the soil, and the total content is no more than a useful guide to the possibility of the occurrence of deficiency or excess of any specific trace-element.

Excesses and deficiencies of some specific trace-elements are still difficult to be caracterised by precise values for the safe limits.

It is known that the plant should satisfy both qualitatively and quantitatively the nutritional requirements of the animals concerned.

The mean daily requirement of the milk-cow for trace-elements is estimated as follows :

1 g	-	Mn
100 - 200mg	-	Cu
1 - 2 mg	-	Co

In order to absorb this quantity of elements, the animals should get a grass mixture containing 50 to 200 mg Mn, 5 to 7 mg Cu and 0.04 to 0.07 mg Co per kg dry-matter (D. M.). In the same way it was stated that the grass should also contain : $\pm$ 26 mg Zn and $\pm$ 12 mg B per kg D. M. Levels above 5 - 10 ppm Mo in the herbage dry-matter must be considered suspect.

Teart in cattle in somerset was reported on pastures with 20 - 100 p. p. m. Mo [LEWIS (1943)].

Several workers have studied the trace element relationship between soil and plant, eventually with regard to the animals, while quite few data have been found concerning the influence of the macronutrients on the uptake of micronutrients by the plant. Considering the influence of macronutrients on the availability and uptake of trace element to the plant, one should distinguish between the originally present and the added macronutrients and observe their ratios.

STEWART and HOLMES (1953) have reported the results of investigations on the micronutrient content of herbage cut repeatedly over a number of years. They did not however separate grasses from clovers, and in consequence any change they found could be due to alternations in the botanical composition of the sward.

Probably more stress should be put on the different factors controlling the trace elements availability to the plant from the soil : the factors existing in the soil, which ultimately will decide upon the plant available fraction.

Such investigation work in different soil conditions and with different plant species for the important elements involved is required.

It would also appear efficient if the discussion should concern the factors affecting the contents of trace-elements in plants in sofar as they influence the health of the animals consuming them.

It may be relevant to mention that for certain elements, such as Mo and Se in the problem of excess is well substantiated. The function of many trace-elements in the plants have been established, but their distribution through the plants is still rather incomplete.

In general the uptake of trace-elements by grasses has been the subject of a great deal of this study. The work reported here is intended as a contribution to demonstrate a clear pattern of the trace-element situation in grasses and clover, as influenced by :

1. - macronutrient fertilizers under the different total conditions.
2. - Organic (Farm Yard Manure) and inorganic (N-P-K) fertilizations.
3. - Trace element nutrition applied in the field and greenhouse conditions.
4. - Contamination along the highway and surrounding industrial areas.
5. - Mobility of some trace elements from soil to plant under

different sources of soil contamination when applied :

1. Chelating agents
2. DTPA, peat and lime.

2. REVIEW OF LITERATURE :

Trace-elements are part of the large number of ions required as a balanced ration for plant nutrition.

The plant and the animal derive the bulk of their supplies of trace-elements from the soil, and eventually from the rock. It is therefore of interest to consider the geochemical distribution of trace-constituents from a magma by the constituent minerals of an igneous-rock which is in process of crystallizing. Concerning the geochemical distribution of trace-constituents in sediments, it seems that many of the correlations found in igneuous rocks, such as that of Nickel with Magnesium, do not persist.

CONNOR et al. (1957) analysed six New-Jersey soils for their total contents of 15 elements, but did not consider the plant availability of any one element.

BEDROSIAN (1965) evaluated the availability to plants of a number of trace elements in New-Jersey soils by means of plant analysis and extraction of the elements from the soil by various procedures. Since the trace element content of grasses and clover may indicate the minerals available to the plants from the soil, the uptake of these elements in an important subject to research from that point of view.

Deficiency and toxicity of trace-elements in plants via the soil and ultimately in the animal nutrition, has been the main consideration in a number of research projects.

The qualitative differences in the trace-element requirements of plants and animals, as well as the considerable quantitative differences which exist both within and between plants and animals, have great practical significance. Deficiencies of Cobalt and Copper are responsible for "steely" wool instead of normally crimped fiber as the product of the sheep's skin (UNDERWOOD, 1956).

A number of workers have studied the distribution of one or more

elements in various plant parts of cereals or herbage plants.
In 1928 BISHOP determined Mn in maize. MILLER (1938) reported 14 major and trace elements in leaves, stems, grain, cobs and roots of nature corn plants grown in 1920. WILLIAMS (1955) dealt with Cu, Zn, Mn, Mo and Fe in subterranean clover at different stages of growth, while FLEMING (1963) gave information for 20 elements in head, leaf, and stem of five constituents sp. of pasture herbage at early maturity. HERMAN, CORNIL and LEDENT (1966) have reported results for 14 elements in rib, heart, green and yellowed leaves of healthy and unhealthy endives. No marked differences between healthy and unhealthy plants were found in the distribution of any element, although the absolute levels varied. WILKINSON and GROSS (1967) have examined the distribution of ten elements in different parts of trifolium repens grown in solution culture.
The effect of fertilizers on herbage composition at specified times has been investigated by such workers as HEMINGWAY (1962), STEWART and HOLMES (1953). WHITEHEAD (1966) has reported valuable information on several aspects of major and trace element uptake by herbage, while observations of WILLIAMS (1959) and FLEMING (1965) are confined to trace elements.
Earlier work on seasonal variation of mineral composition of grasses includes that of BROWN (1943), STEWART and HOLMES (1953), and MELVILLE and SEARS (1953), while THOMAS et al. (1952), BEESON and MCDONALD (1951), VAN PIPER and SMITH (1959) and OYENUGA (1960) are among those who have studied stage of growth effects. DAVEY (1957) has recorded information concerning the distribution of trace-elements in different plant parts at different growth stages.
It is obvious that for several elements there are considerable differences in content from one location to another in the same species.
Variations reported by MITCHELL (1957) are as follows :

Co :	0.07 - 1.5 ppm	in clover and from			< 0.03 - 1 ppm	in rye grass
Mo :	0.21 - 2.4 ppm	"	"	" "	0.3 - 1.5 ppm	" "
Cu :	3.8 - 12.1ppm	"	"	" "	2.4 - 4.3 ppm	" "
Mn :	31.0 - 73.0ppm	"	"	" "	19.0 - 118ppm	" "
Fe :	49.0 - 81.0ppm	"	"	" "	21.0 - 57 ppm	" "
Zn :	20.0 - 40.0ppm	"	"	" "	19.0 - 31 ppm	" "

Deficiencies of mineral nutrition seem to occur more often in legumes than in other crops ; these species are also commonly higher in their concentration of the essential ash elements than other plants, while this group of plants is also high in protein content. This group of plants exhausts the soil more rapidly and severely of its stock of elements others than nitrogen. Lucerne for example reflects the deficiency of boron in the soil by a chlorotic condition in its growing tips earlier and more clear than does any other known field crop (ALBRECHT, et al. 1960).
Several workers have reported figures for different trace-elements deficiencies [PRINCE (1957) and BEDROSIAN (1965)].
The work of CHAPMAN (1966) summarises much information concerning the work being done all over the world in view of determining critical levels of deficiency for different elements in all types of cultivated crops.
The literature however provides no uniform answer to the problem of the relation between trace element content in the growth medium and in the plants.

WILLIAMS et al. (1960) stated that there were no outstanding fertilizer effects on the Cu, Mn or Mo content of clover.
Heavy applications of ammonium sulphate increased the uptake of Cu, Mn and Fe and reduced the amount of Mo, while superphosphate and potash salts were without effect (HEMINGWAY, 1961).
The trace-element content of the plant is nevertheless the primary guide to what the animal recieves and the trace elements most likely to be present in plants in amounts leading to deficiency in animals are : Cu, Co and Iodine (MITCHELL, 1957).
JENCKS (1959) measured vegetable crop yield response to six elements.

Trace elements apparently contribute more to quality than to quantity of nutrition in what we grow, especially the protein (ALBRECHT, 1957). Higher concentration of protein in legume plants, resulting from bacterial inocculation affected more ionic exchange from a standardized soil colloid into the plants.
When by means of trace-elements via the soil, the amino acid nitrogen in the plants can be increased without increasing the total nitrogen, there is a suggestion that the trace-elements may be more fully understood only when we study the plant parts and processes in which the trace-elements probably play their major roles, namely the synthesis of the protein, the enzymes, and the other phases in cell multiplication or cell reproduction (ALBRECHT, 1955).
For such studies, semiquantitative radio-autographic techniques have been applied by different workers [EVANS et al. (1950), ROMNEY and TOTH, 1954), RICEMAN and JONES (1958)].

2. 1. Organic and inorganic sources of trace elements

The application of Farm Yard Manure (F. Y. M.) to the field crops is known as an old practice and F. Y. M. is considered as an important source of trace element supply. It is known also that farm yard manure adds nitrogen, phosphorous, and potassium to a soil and has effect on his physical condition, although some other results observed appeared to be due to some other factors. Several workers : BEAR (1949), PAISLEY (1951), RUSELL (1950) and TURK et al. (1945) have attributed such results to the trace element content. Others have shown that applications of manure have reduced on eliminated disorders due to deficiency of boron [BROWN et al. (1946), DENNIS (1947), HURST (1937), SNYDER et al. (1937)]. Zinc [BARNETTE et al. (1935)]and manganese [SKINNER et al. (1930)] .
Relatively, little has been published on the trace element content of farm yard manure. YOUNG (1935) found a sample of manure to contain 80 ppm of Mn, 50 ppm of boron, and no copper. STEENBJERG (1940) showed that Dannish farm yard manure contained on the average 55 ppm of Mn, 5. 5 ppm of Cu, and 3. 9 ppm of boron. HESTER (1945) found that dry manure contained

15OO ppm of Mn, 35 ppm of B, and 18 ppm of Cu. Other references DENNIS (1947), KATALYMOV (1948), and PAPANOS (195O) have given the boron content of manure at about 2O ppm B.

2. 2. Trace element fertilization :

The importance of trace elements becomes obvious when a deficiency or excess of one or more of these is encountered, and this has been recorded in many parts of the world.
Normally, trace element nutrition is involved to avoid or/and to supply deficiency or correcting toxicity.
Some investigators suggested to raise the concentration of certain elements from sanitary consideration [RADEMACHER (1935), ROSSITER et al. (1948), ANDREWS (1953), MULLER (1964)].
Others are more interested by increasing yearly yields, [GRUHN et al. (1962), KLEINING-LOVEDAY (1962), SMELTZER et al. (1962), KAG-KAGAS et al. (1964)].
MULVEHILL et al. (1955) report that B, Zn, Mn and Cu did not increase yields of Lucerne or oats on 13 different soil types, while yields of an Lucerne, grass mixture were significantly increased by the addition of boron. TRUE and SHREWSBURY (1958) found Mo, Cu and Zn, applied with superphosphate, to be effective in increasing the production of legumes on several texas soils. Applications of those trace elements without superphosphate were ineffective.
FINGER (1951) reports that boron depressed yields of oats, clover and barley, while Cu and Mn increased yields. It also recently was shown that under hungrarian conditions trace element supplementation can raise yields on pastures [HARASZTI et al. (1969)].
In a general way however, it can be put forward, as previously was mentioned by ALBRECHT (1957), that trace-elements apparently contribute more to quality than to quantity of nutrition, unless there are acute deficiencies or toxicities.

2. 3. Interrelations between the major and the trace elements, and between the trace elements themselves.

The uptake of certain elements may be affected by available quantities of other nutrients. For example STOUT et al. (1951)

have found that a large increase in the molybdenum content of plants may be induced through phosphate application, MULDER (1954) has shown an inhibiting effect of manganese on the uptake of molybdenum, and SHIVE (1945) has referred to interrelationships between calcium and boron, and between potassium and boron. When a complex material such as farm yard manure is mixed with soil, there is possibility of a considerable number of interrelationships but it would be very difficult to sort out the effect of one nutrient upon another. MITCHELL (1954) indicated that molybdenum is made more available and that the uptake of boron is reduced by liming. TOTH and ROMNEY (1954) stated that the dominant factor affecting manganese uptake was soil pH. Iron availability within plants is affected not only by pH and by manganese but by the levels of other elements, particularly phosphorus and heavy metals.

DAVIES (1956) referred to a number of papers emphasising the relatinship between lime and molybdenum.

DE KOCK and HALL (1955) found that the P/Fe ratio was higher in chlorotic leaves than in green ones. DE KOCK (1956) working with solution culture and FORSTER (1954) with soil, investigated heavy metals toxicities and found that one effect of these was an induced iron deficiency. WARINGTON (1954) also found interactions between iron and manganese, molybdenum and vanadium at toxic levels.

BROWN (1953) concluded that plants have either a metabolism needing iron or one needing copper. Later, BROWN and HOLMES (1955), and BROWN, HOLMES and SPECHT (1955) investigated the copper-iron and copper-phosphorus-iron relationship in various species.

MULDER (1954) found no copper-molybdenum antagonism in white clover, though some evidence of this antagonism was found by BOLL (1954). OZANNE (1955 a) found that application of nitrogen to white clover increased the severity of Zn deficiency and concluded that this was probably due to retention of more Zn in the roots as Zn protein complexes. KARLSSON (1952) found no effect of phosphorus, potassium, manganese, molybdenum or cobalt on the Zn content of hay.

3. STUDY OF THE INDIVIDUAL ELEMENTS :

3.1. Iron and Manganese : These elements were shown to have a function in photosynthesis [SKOLNIK and GRESISCEVA (1957)], in respiration RUCK and BOLAS (1954) and possibly in translocation (RUCK and BOLAS (1954), SKOLNIK and GRESISCEVA (1957), as well as being concerned in nitrogen reduction.

Nitrogen reduction in plants has been studied in detail, particularly by NICHOLAS and his Co-workers (1957) who showed that the elements Fe, Mn, Cu, Mo and Zn are all involved at some stage.

In animal life, Iron is important particularly as it forms a vital part of the respiratory pigment haemoglobin, and is therefore essential for the functioning of every organ and tissue of the body. Approximately 0.004 % of the total body weight of an animal is iron, of which about 2/3 is to be found in the blood, the remainder being either widely stored in the liver, spleen or kidneys.

Iron was also shown to be in some way concerned with nucleic acid metabolism (KESSLER, 1957) and with amino acids [DEMETRIADES (1956), DE KOCK and MORRISON (1956), (1958 a)] and organic acids [DE KOCK and MORRISON (1958 b)].

Availability to the plants of Mn and Fe is strongly affected by soil pH.

Fe-deficiency can be caused by high soil pH, but lime induced chlorosis does not necessarily mean lower Fe content of the plants suffering from it (ILJIN, 1952 a).

ISLAM and ELAHI (1954) found similarly that waterlogging caused ferric-Fe in soil to be reduced to ferrous, which is more readily available to plants.

Increasing mobility is not always reflected in increased plant uptake : a threefold difference in EDTA-Extractable iron was found between well drained and poorly drained soils.

Mn-content is often related to soil pH. FERGUS (1954) found that a pH 4 French-beans contained 3000 ppm of Mn while healthy plants contained 200-1000 ppm, those with more than 1000 ppm showing toxicity symptoms.

KIPPS (1947) reported that increasing the soil pH to above 6.7

decreased the incidence of Mn-toxicity in lucerne and MORRIS (1948) found sweet clover and lespedeza plants to show toxicity symptoms on soils with high Mn-content and low pH. HASLER and PULVER (1957) reported that plant samples of meadow herbage contained from 18-545 ppm of Mn ; the lowest values were obtained from light soils of the swiss plean, with slightly acid or neutral reaction.

WEHRMANN (1955) found Mn-contents of pasture herbage to be more strongly influenced by soil pH than by soil Mn-content. MCVIKAR (1942) found that Kentucky bluegrass contained more Mn when grown on soils of higher available Mn (i. e. replacable + Mn dioxide). He also concluded that soil aeration is important in Mn uptake, since low aeration would favour reduction to divalent Mn which is more soluble than forms of higher valency.

The effect of liming on mixed pasture and on two of its constituent species showed that liming changed not only their content of Mn, but also their relative proportions in the herbage, increasing clover at the expense of ryegrass. Other factor influencing Mn levels in herbage is soil moisture. Under conditions of impeded drainage, reducing conditions favour the formation of the more mobile forms of Mn, and lead to greater plant uptake. In poorly drained soils the following Mn content has been recorded in different species : ryegrass - 116 ppm ; red clover - 51 ppm.

In well drained parts of the same soil the contents were for ryegrass - 88 ppm Mn and for red clover - 39 ppm : (FLEMING, 1965).

WILLIS and PILAND (1936) found that addition of Cu to soil decreased the availability of both Fe and Mn, probably because of its effect on the redox potential of the soil, which it increased if the soil was aerated and decreased if air was excluded.

The distribution of Iron in the herbage indicates that this element is found mainly in the leaves ; heads contain considerably less and stems least of all. Although variations between the species are wide (FLEMING, 1963) a similar pattern has been observed for the element Mn.

3. 2. Zinc :

Zinc was shown by earlier workers to stimulate the growth of various organism. Probably the first definite evidence for Zn as an essential element was presented in 1914 by MAZE, who demonstrated that without added zinc normal growth of Maize (Zea Mays) was not possible. In 1919 STEINBERG provided proof that Zn is also indispensable for the normal growth of fungi. Later on Zn was definitively considered as an essential micronutrient element [CHASTERS and ROLINSON, 1951 ; GILBERT (1957) ; and HOCH and VALEE (1958)]. Zn plays a role in the synthesis of heteroauxines as it has a regulating function in the formation of tryptophane [SKOOG (1940) ; TSUI (1948)].
KARLSSON (1952) found increases in the Zn content of pasture species after addition of less than 50 kg of Zn sulphate per acre. He also found that addition of Cu, Mn, Mo and Co as well as heavy application of potassium or phosphorous had no effect on the Zn content of plants. Zn is known to be important in the reproduction process within the plant. Evidence of translocation of Zn from leaves in subterranean clover during the production of inflorescences and seeds, has been obtained (RICEMAN, and JONES, 1958).
The concentration of Zn in plants indicated that differences among species do exist, and this can be varied in a wide range [PIPER and WALKLEY (1943, and GLADSTONES (1962)].

Critical concentrations of Zn reported in the literature for the leaves or whole plants of legumes, range mostly between 15 - 20 ppm [TEAKLE and TURTON (1943), VIETS, BOAWN, and CRAWFORD (1954 a & b) ; RICEMAN and JONES (1958)].
Differences in absorption of Zn from the soil and its translocation to the tops, appear to be the main factors responsible for differences among species in susceptibility to Zn deficiency. LOPER and SMITH (1961) found that in bromegrass the Zn content decreased steadily with advance in maturity, but in red and ladino clover the content actually increased.
Application of nitrogen up to 1600 pound per acre increased the Zn content of forage on a dry matter basis (MILLER et al. 1964).
The same author reported that there was an inverse relationship

between the Zn content of the forage and soil pH. Furthermore he found that the Zn uptake per clipping increased with an increase in forage yield.

3.3. Copper :

Cu was shown by NICHOLAS (1957 a & b) to be essential for nitrogen reduction. It is also associated with enzymes, such as terminal oxidases in respiration [ARNON (1950), JAMES (1953), MAPSON (1958)], and propably with the light reaction in photosynthesis [ARNON, 1950].

Another important factor is that of the plant age and seasonal variations : the variation in Cu content was not significant for later stage of maturity or season. Declining Cu content with increase in maturity has been reported by different workers [BEESON and MACDONALD, 1951 ; FLEMING 1965 ; PIPER and BECKWITH, 1949 ; THOMAS et al. 1952].

MITCHELL (1956) did not observe any raise of the Cu-content of perennial ryegrass over 5,9 ppm, even when Cu-sulphate was applied at rate of 60 lb/acre.

THOMAS et al. (1952) quoted the highest value for Cu (15.2 ppm) in the first cut of perennial ryegrass grown in the field.

COOP et al. (1953) also observed that white clover (Trifolium repens) and catsear (Hypochaeris radicata) have higher copper contents than grass species. Similarly DICK et al. (1953) found that the copper content of barley was higher than that of grass.

The increase of uptake and Cu-content by clover over grasses has been commented on by MITCHELL (1956), who found that this occurred especially when soil-Cu levels were high. MULDER (1949) showed that nitrogen application accentuated Cu deficiency on a copper deficient soil. In general application of nitrogen may be expected to accentuate a trace-element deficiency, because the resultant increase in growth will increase the demands which the plant makes on a limited supply, even if there is no direct physiological interaction between nitrogen and the deficient element.

The copper content of pasture plants may or may not show a relation to the total copper in the soil on which they are growing.

LUCAS (1948 b) showed that cupric ions are held tenaciously in organic soils, may be removed by hydrochloric or nitric acids, but not by ammonium acetate. Similar results are reported by several other workers [DICK et al. (1953) ; BECK (1941) ; WILLIAMS (1952) ; MITCHELL (1954) ; and BEESON (1951)]
More recently GOMIDE (1969) observed also a significant decrease in Cu content of forage with plant-age from 4-36 weeks. The same trend occured also for other macro and micro-elements.
HURWITZ (1948) found that the addition of lucerne meal or oat straw to a sandy or silty clay loam, increased the mount of copper leached out by ammonium acetate in proportion to the amount added.
STENBERG et al. (1948) found that total soil analysis was an unreliable means for diagnosing copper-deficiency, but that generally speaking, 20 to 50 ppm of Cu in organic soils, or 8 to 10 ppm in mineral ones, are sufficient for the production of good grass crops with normal Cu-content.
ELLIOT and HUPKENS VAN DER ELST (1956) stated that since Cu is "fixed" in peat soils, responses to Cu-application will go on for some years, fixed in their sense presumably means secure from leaching, but still available to plants. ADAMS and ELPHICK (1956) found the available Cu of several soils to be highly significantly correlated with th e Cu content of white clover growing on them. Soil-pH affected the Cu-content of the herbage, the highest contents occurring on soils with the highest pH-levels WEHRMANN (1955). PIPER and BECKWITH (1951) found a similar effect of pH, but found a larger range of Cu-content between different soils. Cu content in clover at flowering stage, grown in pots with 17 different soils, varied between 1. 8 and 4. 4 ppm. The general pattern is thus similar to that of WEHRMANN (1955) ; but PACK et al. (1953) found no dependable relationship between soil pH and Cu content of crops. Also in some Yougoslavian soils the relationship between soil-pH and soil-Cu content has been observed, a decrease in Cu content being accompanied by an increase in pH (ZAGORKA et al., 1960). Considering plant-soil relationship, TEAKLE et al. (1942) found that subterranean clover could be used as an indicator for the copper status of the soil ; they found low Cu-contents on sands and

gravelly sands as did UNDERWOOD et al. (1943), and much higher figures on loams.
CUNNINGHAM (1950) stated that most of the New-Zealand's pastures deficient in Cu, occur on coastal sands, leached silts of peats. BECK and BENNETTS (1942) and BECK (1941) reported a pronounced effect of soil type.
On the other hand PIPER and WALKLEY (1943), while indicating a possible correlation of the Cu-content of oats, stated that Cu in plants is more affected by species than by soil type.
In recent years the Aspergillus Niger method has been used by several workers as a means of assessing the amount of soil Cu available to plants, and in some cases the values obtained have been correlated with the incidence of Cu deficiency in livestock. Thus ACOCK (1941) suggested that 2 ppm in the soil is the treshold figure, below which pastures are likely to be deficient in Cu. WRIGHT (1945) quoted for New-Zealand soils 1.5 ppm as the minimum safe level for mineral soils.
VANDERELST et al. (1961) and HALLSWORTH et al. (1960) suggested that Cu is also concerned with the nitrogen fixation.

3.4. Boron :

The essential nature of boron was first suggested by AGHULON (1910), and later by MAZE (1951), (1919) especially for corn. This discovery was confirmed beyond any doubt for broad been (Vicia faba) by the work of WARINGTON (1923). The effect of boron deficiency on different plants has been investigated by several workers : DENNIS et al. (1939) ; (1941) ; (1943) ; DENNIS (1937) ; (1937) ; LOHNIS (1940) ; BRANDENBURG (1939) ; JAMALÄINEN (1936) and CHANDLER (1941).
The problem of boron deficiency is most pronounced in humid regions, since the available boron is rapidly lost by leaching. While boron excess mainly occurs in saline soils also acid soils without leaching may contain high amounts of available boron.
Boron seems to be an essential and important element for man, plants and animals (HOVE, 1939). It has been recommended that all mixed fertilizers should contain 0.025 % B. Additional amounts

of boron are suggested for those crops, which requirement for this element is high, for example Lucerne, tomatoes, celery and some root crops (REEVE et al., 1948 ; WOLF, 1940).
MACLEAN and LANGILLE (1958) found a direct correlation between available soil boron and that in lucerne tissue. POWER and JORDAN (1950), claimed that the level of available boron in the soil should be 0.5-1.0 ppm under which conditions lucerne should contain 20-50 ppm.
STINSON (1953) found boron deficiency in lucerne if plants contained less than 20 ppm. This occurred on heavy soils with less than 0.5 ppm water soluble boron, or on sandy soils with less than 0.3 ppm. He also found that moisture deficiency tended to increase development of boron deficiency symptoms. DIBLE and BERGER (1952) found that the boron content of the upper parts of lucerne plants decreased as soil moisture became limiting in the surface 12 inches. WARING (1956) however, found boron deficiency on waterlogged soil on which it did not otherwise occur. The concentration of boron in the tips of leaves with parallel veins has been investigated by KOHL and OERTLI (Unpublished data) who concluded that a physical concentration of boron occurs in the tissue water which flows toward the tip due to transpiration. Such a theory can explain the increasing boron concentration from base to tip and it could also explain why the boron concentration increases with increasing age of the leaf.
Soil texture has some effect on the incidence of boron deficiency ; KUBOTE et al. (1948) showed that most applied boron moved to 24 inches or deeper in six months on soils of light texture, but much of it was found at 12 inches or less on heavier soils. The results of WILSON et al. (1951) agree with this. ANDERSON (1952) obtained good responses from lucerne and subterranean clover towards 3.5 lb of boron per acre, and residual effects of this element were still evident after six years. On the other hand, MCGREGOR and MALVEHILL (1955) found that 20 or 30 lb of boron gave a significant increase in the boron content of lucerne and oats in the year of application, but not in the following one. However, there was no evidence of deficiency, since borax application had no effect on yield.

WEAR (1956) found the greatest increase in seed production by crimson clover, following an application of 10 lb of borax per acre, and there was no toxic accumulation in sensitive species during four years after applying 30 lb per acre. JAMALAINEN (1950) reported that the main problem with boron deficiency in Finland is that of correcting soil acidity without causing boron deficiency through excess of added calcium.

It appears that tolerant species must accumulate boron at a slow rate. The concentration of boron in the transpiration stream in localized areas, may serve to explain why there is a relatively narrow range between deficient and toxic boron concentration in the solution (DIBLE & BERGER, 1952). Boron toxicity to plants is a matter of particular concern in arid regions of the world, where this element is often present in damaging concentrations in irrigation waters (OERTLI and LUNT et al., 1961). The comparison between tolerant and more sensitive grasses, favours the hypothesis that the sensitivity towards excessive boron supply is related more to uptake rates than to differences in tolerance of tissues (OERTLI, et al. 1961). SKOLINIK and SOLOVIYOVA (1961) disagreed with GAUCH and DUGGER (1953) on the cause of root tip death under conditions of boron deficiency, which they attributed to distrubance in nucleic acid metabolism.

3.5. Molybdenum

Molybdenum differs from most other trace-elements in that its availability to plants is increased with increasing pH. LEWIS (1943) shows in his work on the "teart" pastures of Somerset how this principle operates. The plants were found to have maximum molybdenum when the pH was between 7 and 8. BARSHAD (1951) found that the uptake of Mo by ladino clover, birdsfoot trefoil, rhodes grass and ryegrass from a clay loam soil was suppressed when the soil was acidified with sulphuric acid. Others found that a change in the soil reaction from pH 4.8 to 7.3 increased the uptake by Medicago denticulata, Erodium Cygnorum and Hordeum leporium, while alkalinities beoyond pH 8 produced higher increase in the uptake of Mo. Under conditions of low Mo supply, grass may have a higher level than associated clover, but when the supply is abundant the reverse is generally the case (MITCHELL, et al. 1957).

The phenomenon known as "teart" in pastures has been recognised as a severe case of Mo excess for both plants and animals. It has been shown that teart herbage was very much higher in Mo-content than non-teart herbage. The role of Mo in nitrogen fixation and nitrate assimilation has been further studied for legumes (MULDER et al., 1959 a & b), and for non legumes (BOND & HEWITT, 1961 ; BECKING, 1951). MINIMA (1960) as well as STEWART and MARGOLIS (1962 b) have suggested that Mo might be concerned in amino-acid metabolism. Another significant point related to Mo is that the range of contents within which normal plant growth occurs, is extremely wide. On the other hand it is toxic to ruminant animals in quite small concentrations. ASKEW (1956 a) considers that on soils low in calcium and molybdenum, application of Mo may so increase growth, that Ca is seriously depleted and vice versa. ASKEW (1956 a) also found that application of lime and molybdenum reduced the levels of Mn in pasture plants in the autumn. Mo content of Subterranean clover was found by PIPER and BECKWITH (1951) to decrease, and by BARSHAD (1951) to increase with age. BARSHAD claimed also that for any particular species Mo content varies inversely with growth rate, so that it tends to increase particularly during periods of slow growth. Presumably this is due to the fact that growth rate is more directly affected by external factors than Mo uptake.
Mo-content of pastures is more closely related to availability of soil-Mo than the total quantities present. DAVIES (1956) has discussed the factors affecting availability and lists them as :

1. pH : as this increases, availability increases.
2. Phosphate supply : availability increases with increase in phosphate, probably because of increased translocation of Mo.
3. Sulphate and Mn : both of which depressed Mo-uptake.
4. And K, with which there is a positive interaction.

In general alkaline, organic and some young soils (e. g. Volcanic) tend to have high Mo contents, while podsolised soils,

calcareous sands and serpentine soils tend to have absolute deficiencies.
Soils of high anion exchange capacity and low pH, iron-stone soils, and depleted soils have low available Mo. Liming has also often given responses on ironstone and acid soils (DAVIS et al., 1951; EVANS et al., 1951 ; MCLACHLAN, 1955 ; ROBINSON et al., 1951 ; ROBINSON and EDIGNTON, 1954 ; ROSSITER, 1952 ; STEPHENS and OERTEL, 1943 ; OERTEL et al., 1946 ; PIPER and BECKWITH, 1951 ; STOUT et al., 1951 ; WALKER et al., 1955 ; EVANS et al. (1951) found that the Mo-content of lucerne was increased from O. 7-1. 1 ppm to 1. 6-3. 3 ppm, by liming the soil to increase its pH from 6. 1-6. 5 to 6. 9-7. 6 ppm.
ROBINSON et al. (1951) found uptake by ryegrass and several legumes to be increased 6 to 11 fold by liming to pH 7.
ROSSITER (1952) found a negative interaction between lime and Mo on a sandy soil of pH 6. 1, but this varied with time of application and K level. He observed that nitrogen is primary concerned in responses to lime and Mo, and that Mo concentration in the plant gives little indication of the needs for good growth. He suggested that lime is mainly acting on the transport of Mo from roots to tops of plants.
MCLACHLAN (1955) found that Mo-deficiency decreased with increasing soil pH. He also found no correlation between the occurrence or intensity of deficiency and geological origin of soil, climate, soil colour, texture or organic matter content.
All this confirms the close relationship between soil pH and plant Mo content, while also some other interactions have been observed.

3. 6. Cobalt

Cobalt has been proved to be essential for plants. However MILLER (1954) and THIMANN (1956) have shown that it increases the growth rate of etiolated pea stems, while BOLLE-JONES et al. (1957) observed beneficial effects on the growth of rubber plants. BOND and HEWITT (1962) have shown Co to be essential for the nitrogen fixing symbionts of casurina and alnus. SCHARRER and TAUBEL (1954), found that application of complete N-P-K fertilizer increased the uptake of Co, which was toxic at high rates of application (25 kg Co-Chloride per hectare). Application of boron appeared to favour the uptake of Co, but that of Fe or Mo depressed it. WEHRMANN (1955) found the Co-content of herbage to vary directly with the soil content, though it was also affected by soil pH. RIGG (1940) stated that there is no direct connection between the cobalt content of the soil and this of plants, but that there is apparently a close connection between cobalt in grass and soil pH. From the point of view of the animals it can be mentioned that Co is a constituent of vitamin B_{12}, which is essential for the nutrition of some and possibly all animals. Sheep suffer from deficiency when their food contains less than 0.07 ppm Co and cattle when it contain less than 0.04 ppm. Parti cle size fraction is one of the factors affecting the Co status of the soil. Co-content increases with decrease in particle size with depth within the soil (CLYDE, 1953). The clay fraction contains about seven times as much Co than the sand.

Plants from poorly drained soils were higher in Co than were these from well drained soils. Applications of liming materials reduced availability of soil Co.

Lucerne from soil with pH values higher than 7.2 contained only about half as much Co as from soils with pH values of 5.8.

An increase of the amount of available Mn or Fe in a nutrient solution caused a marked reduction in the amount of Co taken up by the plant.

The Co-content of various plant species when grown under the same conditions ranged between 0.01-0.70 ppm. In general, legumes, cereal forages, kentucky bluegrass forage and most weeds are relatively high in Co. Mature cereals grasses and underground portions of vegetables are relatively low in Co.

CHAPTER II

SPECIFIC INFLUENCE OF VARYING LEVELS OF MACRO ELEMENTS ON TRACE ELEMENT UPTAKE BY PERENNIAL RYEGRASS

1. INTRODUCTION

Since the main purpose of the present thesis is to study the influence of macroelement fertilizers on the uptake of trace elements by pasture crops, it was necessary to separate the macronutrient (i. e. N, P, K, Ca and Mg) applications, in order to study their specific effects on the minor element status.
Different levels of major elements were low, medium, high and very high for each element. Results of the present experiment indicate the specific interaction between each level of the major element applied and the minor elements in the plant tissues, as well on the yields of dry matter as on the uptake.

2. EXPERIMENTAL DETAILS

A sandy loam soil collected in 1968 on a control plot (without fertilization since 1959) of a fertilization trial at a depth of O-15 cm was used. The soil was dried, crushed and potted in plastic pots containing 1 kg each. The N, P, K, Ca and Mg levels applied were as follows :

Table 1 : Amounts of fertilisers added as kg/ha

	Treatments in units per ha						With basic addition kg/ha		
Nitrogen as NH_4NO_3	N_0 0	N_1 25	N_2 50	N_3 100	N_4 200	N_5 400	NH_4NO_3 -	$Ca(H_2PO_4)_2 2H_2O$ 40	K_2SO_4 100
Phosphorus as $Ca(H_2PO_4)_2 2H_2O$	P_0 0	P_1 10	P_2 20	P_3 40	P_4 80	-	100	-	100
Potassium as K_2SO_4	K_0 0	K_1 25	K_2 50	K_3 100	K_4 200	K_5 400	100	40	-
Calcium as $CaCO_3$	Ca_0 0	Ca_1 500	Ca_2 1000	Ca_3 2000	Ca_4 4000	-	100	40	100
Magnesium as $MgSO_4$	Mg_0 0	Mg_1 25	Mg_2 50	Mg_3 100	Mg_4 200	-	100	40	100

Each treatment was replicated four times and each replication, representing 1 kg of soil, was mixed separately in a plastic container. All treatments were added in a sufficient quantity of solution to bring the soil to field capacity. After mixing and potting, 75 seeds of Lolium perenne, L. Perennial ryegrass (pasture type C. V. Vigor) were sown in each pot ; the layout was completely randomized, and successive cuttings were harvested every 25 days. Soil moisture was kept throughout the whole experiment at field capacity with deionized water.
Greenhouse temperature was $\pm$ 15° C and the plants were given artificial fluorescent light for 16 hours per day.

2. 1. Soil used

As far as the soil classification is concerned, all the soils reported under the present study were classified according to the Belgian classification.
The soil with pH KCl = 5. 1O and pH H_2O = 6. 3O was found to show the following characteristics :

Mechanical analysis :

Sand	5O - 67. 5 %
Silt	2O - 5O %
Clay	$<$ 12 %

Chemical analysis :

C - 2. 3 g/ 100 g of dry soil (WALKLEY & BLACK)
CEC - 11. 2 meq/ 100 g soil - (MEHLICH method)

Ca	- 4. 8O meq/ 100 g soil	extracted with NH_4Ac - pH 4. 8
Mg	- 0. 43 meq/ 100 g soil	
K	- 0. 20 meq/ 100 g soil	
Na	- 0. 11 meq/ 100 g soil	
P	- 1. 18 mg/ 100 g soil	

The trace element status was determined by extraction with O. 1 and O. 5 N HNO_3.

Extractable trace elements (ppm in air dry sample)

	Fe	Mn	Al	Zn	Cu	Pb	Mo	Co	Ni
0. 1 N HNO_3	162	42. 5	225	10. 50	2. 75	5. 50	tr.	1	1
0. 5 N HNO_3	1200	105	437. 5	17. 25	17. 25	19. 50	tr.	tr.	4

Statistical analysis :

In the present experiment, the data are the concentration and the uptake of trace elements, Fe, Mn, Zn, Cu, B and Pb. The treatments being quantitative, the analysis was considered as a regression problem. For each trace-element and for each cut, an analysis of variance was made. In this analysis the sum of squares of treatments is subdivided into a linear, quadratic, cubic and a rest component. These components (except the last one) were tested against the residual error. If one (or more) of them was significant, the regression was calculated.

3. RESULTS AND DISCUSSION

3. 1. Plant growth and dry matter yields

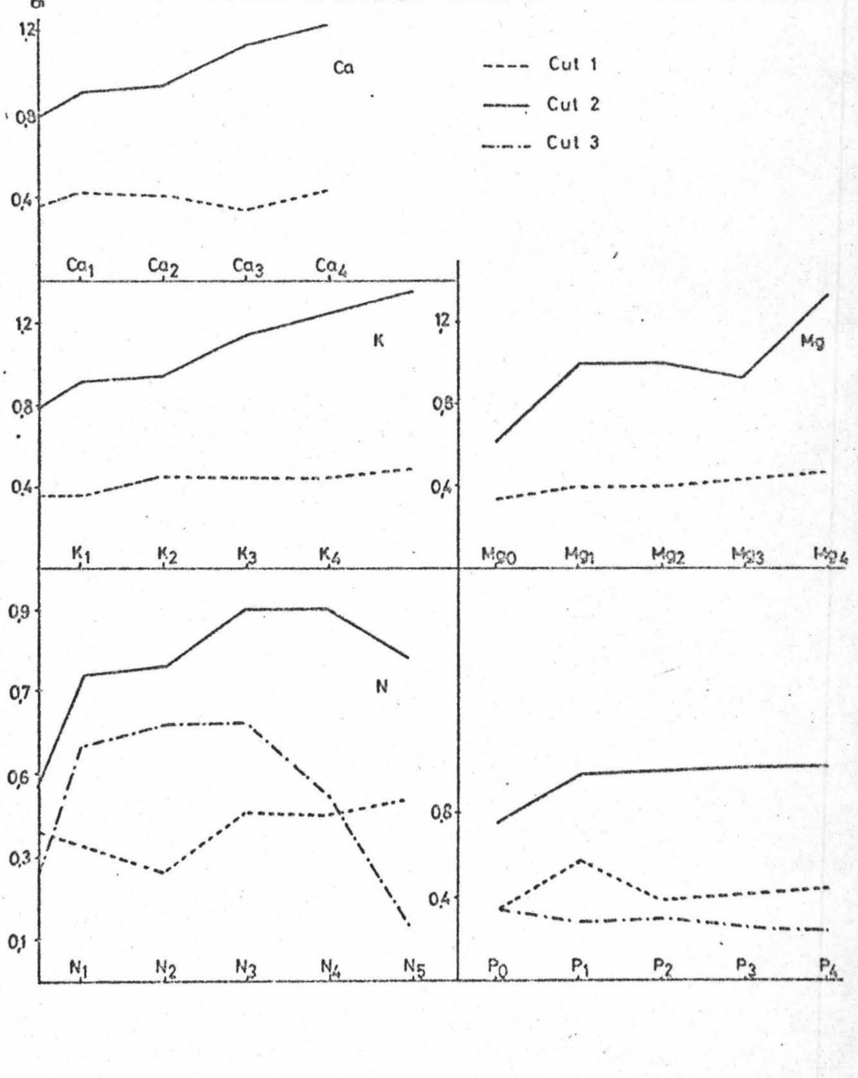
From the nitrogen and phosphorous treatments three cuttings were obtained, from the potassium, calcium and magnesium treatments only two.

In a general way the first cuts did not show the same quantitative response as the following ones. This is an indication for the fact that the fertilizers used were not acting immediately but only after a certain time of integration in the soil. It may be added that the highest yields were generally obtained during the second growth period.

3. 1. 1. Effect of Nitrogen

The main effect of N on the dry matter yield appeared in the second and third cuts. Since a marked decrease was observed at the rates N_4 and N_5, the yield curves of these two cuttings showed the classic form of an optimum curve. The first harvest however, did not follow the same trend and gave less variation in function of nitrogen applications (Fig. 1.).

FIG. 1 DRY MATTER (g/kg of soil) YIELDS OF PERENNIAL RYEGRASS SHOOTS AS INFLUENCED BY DIFFERENT LEVELS OF MAJOR ELEMENT FERTILIZATION



3.1.2. Effect of phosphorus

There was no systematic nor significant influence of the phosphate treatments on the yields obtained. This confirms the fact that the soil being used was sufficiently rich in phosphorus.

3.1.3. Effect of K, Ca and Mg

For each of these elements, increasing applications resulted progressively increasing yields of the second cut.

3.2. Trace element uptake

The results of the trace element concentration and uptake are illustrated in figures 2 to 11.

3.2.1. Effect of Nitrogen

The iron content shows that there were no appreciable effects of nitrogen levels on the iron concentration in the plants (table 2). The second cut tended to accumulate higher values of iron than the others.

The mean values of three cuts at N_1 and N_5 were 297 and 290 ppm respectively. Results reported by HEMINGWAY (1962) indicate also that there is no evidence to suggest that nitrogen fertilisations reduce the level of iron, but he reported an increase of Fe absorption towards macronutrient fertilizers such as ammonium sulphate while GOMIDE et al. (1969) found no such an effect on tropical grasses. It may be relevant to mention that the extractable amount of iron in the soil was found to be four times the amount of Mn extracted with 0.1 N HNO_3, and still more where extracted with 0.5 N HNO_3, though the manganese concentration and uptake by the grass wasmuch higher than that of Fe. This shows that availability is the limiting factor rather than soil content, and numerous factors have been shown to be concerned with this.

The Fe/Mn ratios found at different levels of nitrogen application in the different cuts were as follows :

Cut	Control	N_1	N_2	N_3	N_4	N_5
1	-	1:7	1:6	1:7	1:7	1:7
2	1:4	1:3	1:10	1:7	1:6	1:4
3	1:5	1:8	1:6	1:8	1:2	1:6

In fact both curves, of yield production and iron absorption, revealed a parallel relationship. Thus the maximum yield and maximum Fe uptake correspond with the rate N_3.

Table 2 : Statistical differences in trace element concentration (p.p.m. in dry matter) and uptake (μg/pot) as affected by addition of ammonium nitrate.

		Concentration						Uptake					
Regression	Cut	Fe	Mn	Zn	Cu	B	Pb	Fe	Mn	Zn	Cu	B	Pb
Linear	1	N.S.	N.S.	*	**	**	*	N.S.	*	N.S.	N.S.	**	N.S.
	2	*	**	N.S.	**	*	N.S.	N.S.	*	N.S.	N.S.	N.S.	N.S.
	3	N.S.	*	N.S.	*	**	*	*	N.S.	N.S.	**	**	**
	Total	N.S.	**	*	**	**	N.S.	N.S.	*	N.S.	**	N.S.	N.S.
Quadratic	1	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	*	N.S.	N.S.	N.S.	N.S.
	2	**	**	N.S.	**	**	N.S.	N.S.	**	N.S.	N.S.	**	N.S.
	3	N.S.	**	N.S.	**	**	N.S.	**	N.S.	*	N.S.	N.S.	N.S.
	Total	*	**	N.S.	**	**	N.S.	N.S.	N.S.	*	N.S.	**	N.S.
Cubic	1	N.S.	N.S.	N.S.	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	**	N.S.
	2	**	**	N.S.	N.S.	**	N.S.	N.S.	**	N.S.	N.S.	N.S.	N.S.
	3	N.S.	N.S.	*	**	*	*	*	N.S.	**	*	N.S.	N.S.
	Total	N.S.	**	N.S.	*	**	N.S.	N.S.	**	*	N.S.	N.S.	N.S.

* significant at 5 % ; ** significant at 1 % ; N.S. not significant

According to MCVICKAR (1942) the soil parent material is not necessarily an index of the manganese content of the soil, but STEPHENS (1951) found that mineral deficiencies occur in Australia almost entirely on three groups of soils with particular genetic features. It is noteworthy that under the present conditions the plants absorbed and accumulated high quantities of manganese, without exhibiting toxicity symptoms.

Different authors have mentioned Mn concentrations of 1000 ppm and more in plants growing under conditions of high acidity and high manganese supply. FERGUS (1954) found that at pH 4 French beans contained 3000 p. p. m. of Mn, while healthy plants contained only 200-1000 p. p. m. ; those with more than 1000 p. p. m. showed toxicity symptoms. In this case, the pH of the soil, being 5. 7, is such that the uptake of manganese was certainly favoured.

It is known that the manganese content of herbage is profoundly affected by changes in soil reaction : soil pH has a stronger influence than its Mn content (WEHRMANN 1955). Manganese is one of the factors concerned in the soil-acidity complex (SCHMELL et al. 1950 ; HALLSWORTH et al. 1957), and resistance to injury varies considerably between plant species and varieties (DESSUREAUX and ONELETTE, 1958 ; TURCIN and SOKOLOV, 1950).

It appears that it is not sufficient to explain the high increase in manganese absorption by the soil pH factor, since the soil pH was uniform for all the treatments (see under 2. 1.); the high Mn accumulation and absorption seem to be related to other factors which are not immediately apparent.

From the literature different effects of nitrogen on Mn were reported ; GOMIDE et al. (1969) found an increase in Mn with N fertilization, while earlier WILLIAMS et al. (1960) observed no such an outstuding effect. Results of the current experiment showed that high manganese concentrations were generally obtained, but the fluctuations occuring between the different nitrogen treatments did not follow a uniform pattern.

There was a sharp reduction in Mn absorption, at the highest rates of nitrogen i. e. N_4 and N_5 in the second cut.

Therefore with regard to the different nitrogen levels and over all the cuts, the manganese concentrations resulted in a significant quadratic regression (table 2). Concerning the total Mn uptake there is a clear tendency to decrease towards the last cut. This means that the second cut had the maximum values of manganese uptake for all the treatments, while the highest Mn absorption occured at the rate N_3.
Irrespective of the fluctuation pattern obtained in relation to the different cuttings and increasing rates of nitrogen, the high Mn availability may also be influenced by the iron content of the soil. BOKEN (1956, 1957) observed that ferrous salts could reduce Mn to the divalent form in which it is more readily available to plants. As already mentioned the present soil values indicated a high Fe level compared to Mn.

The Zn concentrations found were situated between 76 ppm and 176 ppm and the zinc content showed a certain decrease with increasing rates of nitrogen. This may be due to a dilution effect occurring with increasing yields of dry matter.
It is also observed that the Zn content of the grass was increasing in the successive cuts, but different nitrogen levels had no systematic influence.

There was no outstanding effect of nitrogen levels on the zinc uptake, during the first cut. During the second cut and the third cut this effect was more pronounced and at the level N_3 the maximum zinc absorption was corresponding once again with the maximum yields. This explains why Zn uptake varied between 15 and 124 microgram Zn per pot (fig. 3).

The soil being used has a quite normal content of Copper, which was 5.7 ppm extractable with O.5 N HNO_3.
The copper values found in the plants of this experiment ranged between 24 and 6O ppm, the highest values being systematically found in the second cut. Comparing our results to BEESONS' (1947) classification all the contents belong to the high level (more than 12 ppm).
The critical value of copper content in grasses is generally quoted as 7 to 8 ppm with relation to animal nutrition.

FIG. 2 INFLUENCE OF DIFFERENT LEVELS OF NITROGEN ON TRACE ELEMENT CONCENTRATION (P.P.m in dry matter) BY SHOOTS OF PERENNIAL RYEGRASS

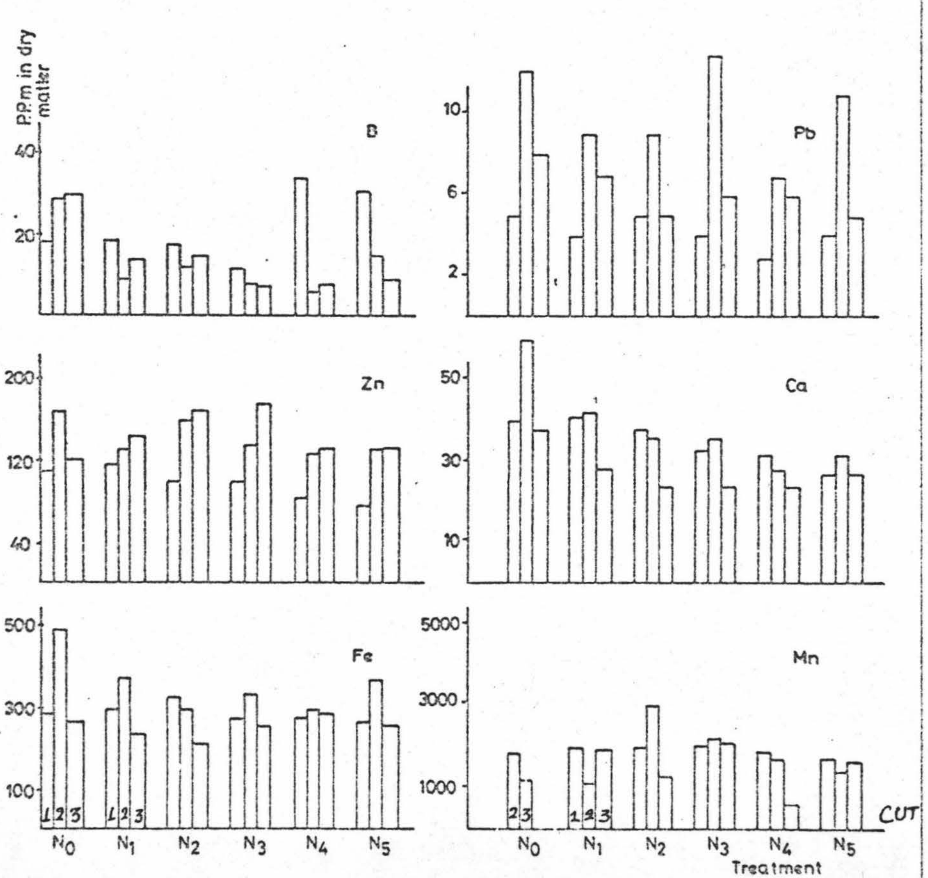
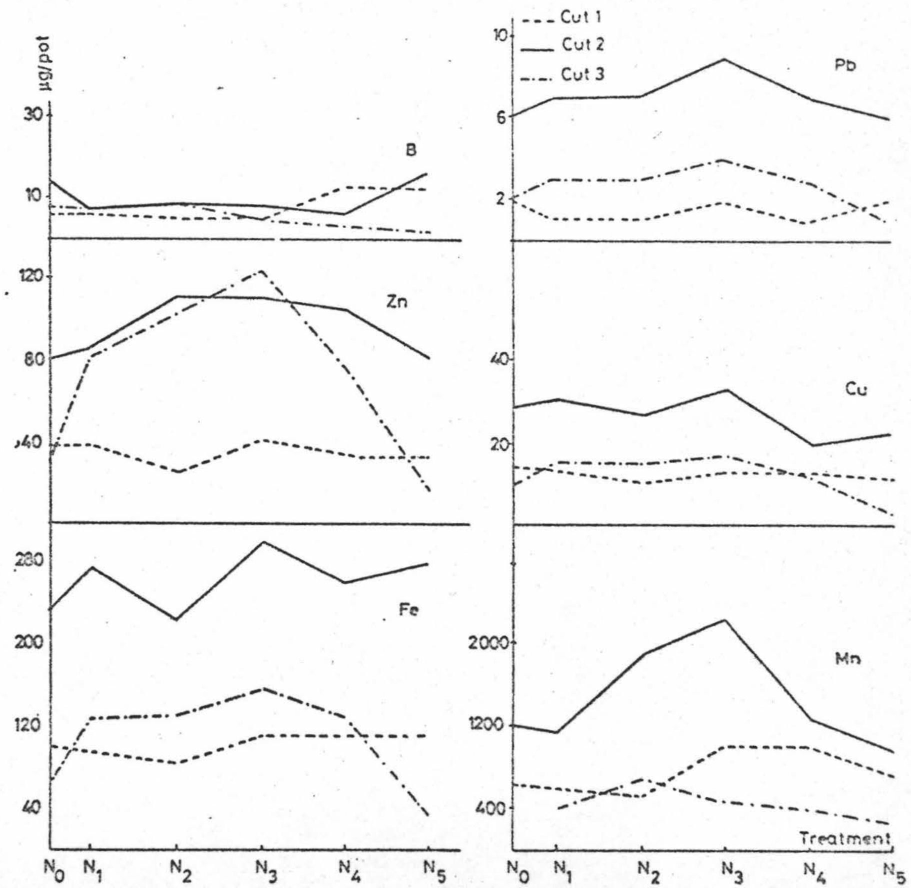


FIG. 3 INFLUENCE OF DIFFERENT LEVELS OF NITROGEN ON TRACE ELEMENTS UPTAKE (μg/ pot) BY SHOOTS OF PERENNIAL RYEGRASS



This corresponds with the highest content found by WEHRMANN (1955) in varying soil conditions. The high values found in the present experiment could be explained by the high density of roots developing in the pots with one kilogram of soil and resulting in an intensive utilization of the available quantities.
Some authors observed a change in soil pH as a results of major element application, but the influence of pH on copper uptake is still a matter of contro verse (PIPER and BECKWITH 1951, PACK 1953). In the present case the slight change in soil pH caused by the nitrogen treatments could not be responsible for differences in copper absorption. Considering the total uptake, the general trend observed indicates a certain decrease with increasing nitrogen application.

The boron content of the harvested plants ranged between 6 and and 34 p. p. m. This element is known as being very variable in the plants, and the variations observed here are relatively small. Boron is the only element for which the first cut gave the highest concentration in the dry matter for all nitrogen levels. Another difference in comparison to the other elements is the fact that the total uptake of boron was quite similar for each of the three cuttings.

3.2.2. Effect of phosphorus

In spite of the fact that some authors observed antagonistic effects of phosphate with respect to trace element uptake (BROWN et al. 1959, DE KOCK et al. 1955), our increasing treatments with PO_4^{---} did not permit to observe any systematic decrease of trace element absorption. The existance of positive correlations between phosphorus and microelements (Fe, Mn, Cu, Co and Zn) has been stated by LOPER and SMITH (1961) in lucerne, red and white clover and smooth bromegrass.
HANLEY (1962) observed relatively small variations in boron content of ryegrass in function of varying levels of phosphate.
The trace element variations caused by our different treatments are of the same order as the ones observed between the successive cuttings. Except for boron, the total uptake of all the trace elements was higher in the second cut than in the other two. As we have men-

FIG. 4 INFLUENCE OF DIFFERENT LEVELS OF PHOSPHORUS ON TRACE ELEMENT CONCENTRATION (P.P.m in dry matter) BY SHOOTS OF PERENNIAL RYEGRASS

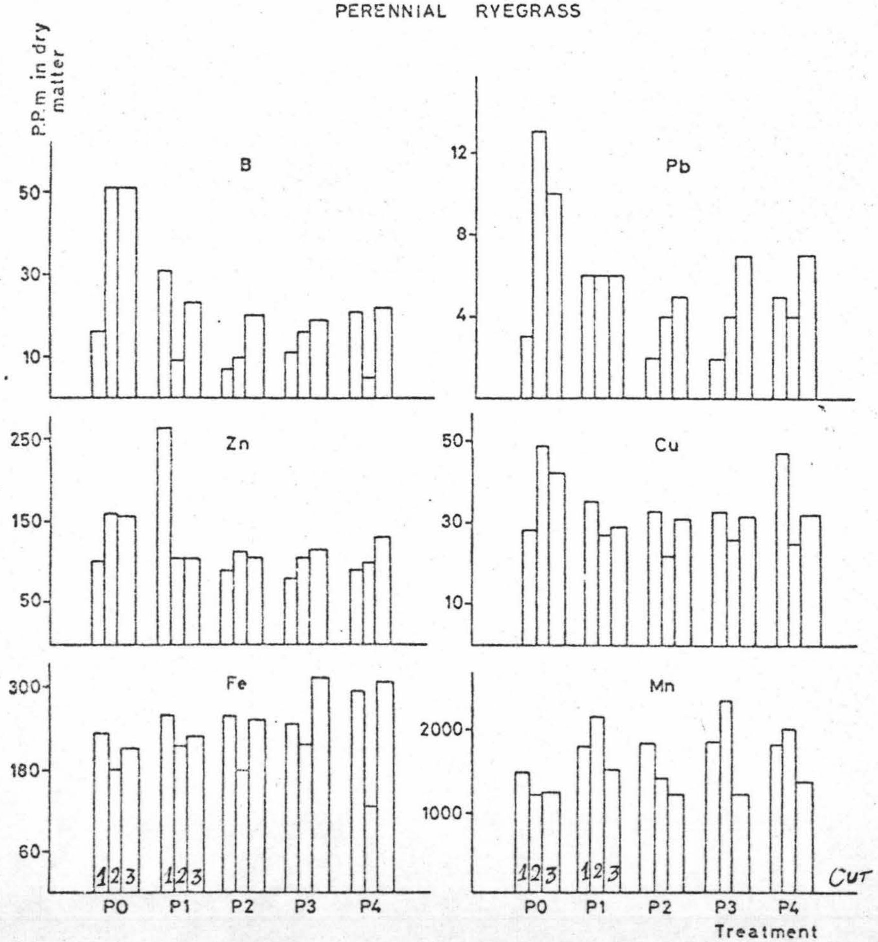


FIG. 5 INFLUENCE OF DIFFERENT LEVELS OF PHOSPHORUS ON TRACE ELEMENT UPTAKE (μg / pot) BY SHOOTS OF PERENNIAL RYEGRASS

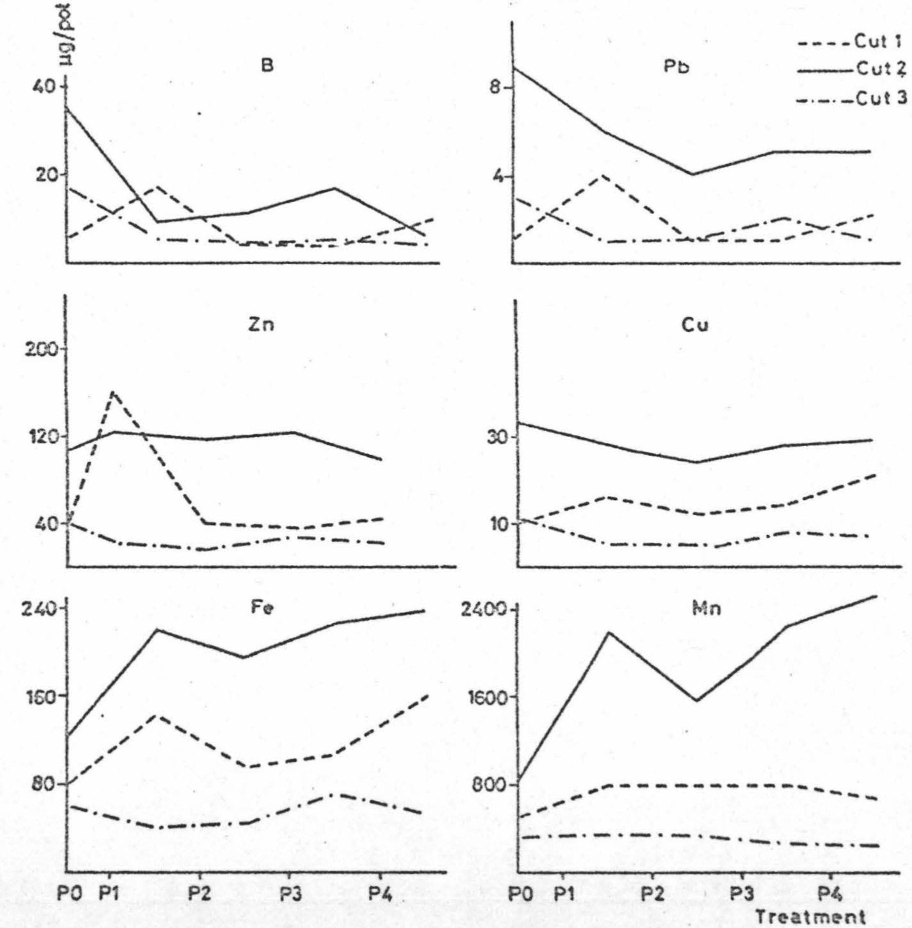


Table 3 : Statistical evaluation of trace element concentration (p.p.m. in dry matter and uptake (μg/pot) as affected by addition of mono calcium phosphate

		Concentration						Uptake					
Regression	Cut	Fe	Mn	Zn	Cu	B	Pb	Fe	Mn	Zn	Cu	B	Pb
Linear	1	*	*	**	**	N.S.	N.S.	*	N.S.	N.S.	*	N.S.	N.S.
	2	N.S.	**	*	**	**	*	**	**	N.S.	N.S.	**	N.S.
	3	**	N.S.	N.S.	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	**	*
	Total	**	*	**	N.S.	**	*	**	**	N.S.	N.S.	**	*
Quadratic	1	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
	2	N.S.	**	N.S.	**	**	*	*	**	N.S.	N.S.	N.S.	N.S.
	3	N.S.	N.S.	*	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	**	N.S.
	Total	N.S.	**	**	**	**	**	N.S.	**	N.S.	N.S.	**	N.S.
Cubic	1	N.S.	*	**	N.S.	*	**	N.S.	N.S.	*	N.S.	*	**
	2	N.S.	N.S.	N.S.	**	**	N.S.	N.S.	N.S.	*	N.S.	**	N.S.
	3	N.S.	N.S.	N.S.	N.S.	**	*	N.S.	N.S.	N.S.	N.S.	**	N.S.
	Total	N.S.	N.S.	N.S.	**	**	N.S.	*	*	N.S.	N.S.	*	N.S.

* significant at 5 % ; ** significant at 1 % ; N.S. not significant

tioned already, the soil being used was not deficient in phosphorus, so that the effect of phosphorus treatments may have been attenuated by its original level. However, table 3 shows several significant regressions between phosphorus levels and concentration and uptake of trace elements. The elements Zn and Cu gave less significant response than most of the other ones (Table 3).

3.2.3. Effect of potassium

There was a quite pronounced effect of potassium treatment on the content of some trace elements in the plants. The general trend was a decrease, especially during the second growth period, and the elements boron and copper gave the clearest response. In this particular case the same was also observed for lead (table 4).
At the highest potassium level, the boron concentration in the first cut dropped down till the limit of detection and for this element the difference between first and second cut was the highest. It may be mentioned that DIBLE and BERGER (1952) considered the 9 ppm B level in lucerne as the limit below which deficiency might exist.
The elements Zn, and Mn were quantitatively less sensitive towards potassium treatments.
The trace content of the plants was higher in the second cut than in the first for all elements except Mn. The ratio Fe/Mn was of the order of 1:10.
As both Mn and Fe were present at levels, higher than 100 ppm, the phenomenon of iron-manganese antagonism or competition must be acting here.
The total uptake of trace elements was also the highest during the second cut. Differences in function of K-treatments were however more levelled and less frequent (Fig. 7).

3.2.4. Effect of Calcium

Effect of Ca on the soil

Due to the fact that Calcium, applied as $CaCO_3$ altered the pH of the soil, this factor gave the clearest response with respect to trace element uptake.

FIG. 6 INFLUENCE OF DIFFERENT LEVELS OF POTASSIUM ON TRACE ELEMENT CONCENTRATIONS (P.P.m in dry matter) BY SHOOTS OF PERENNIAL RYEGRASS

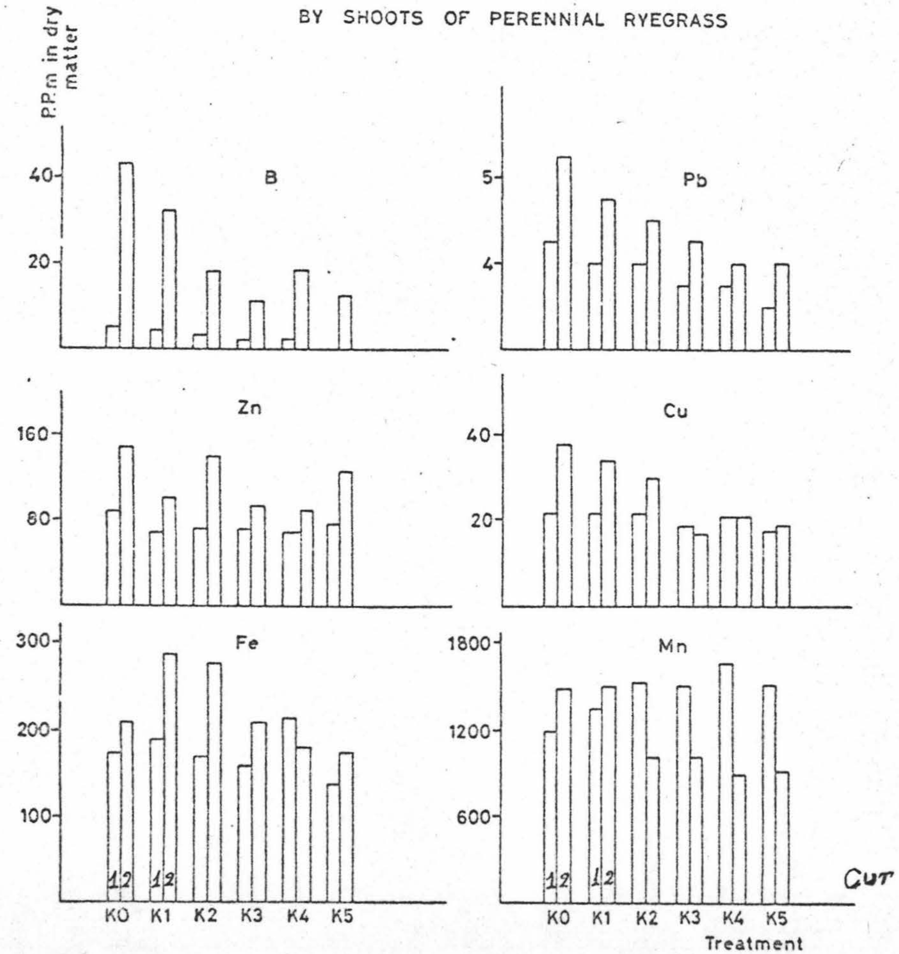


FIG. 7 INFLUENCE OF DIFFERENT LEVELS OF POTASSIUM ON TRACE ELEMENT UPTAKE (µg pot) BY SHOOTS OF PERENNIAL RYEGRASS

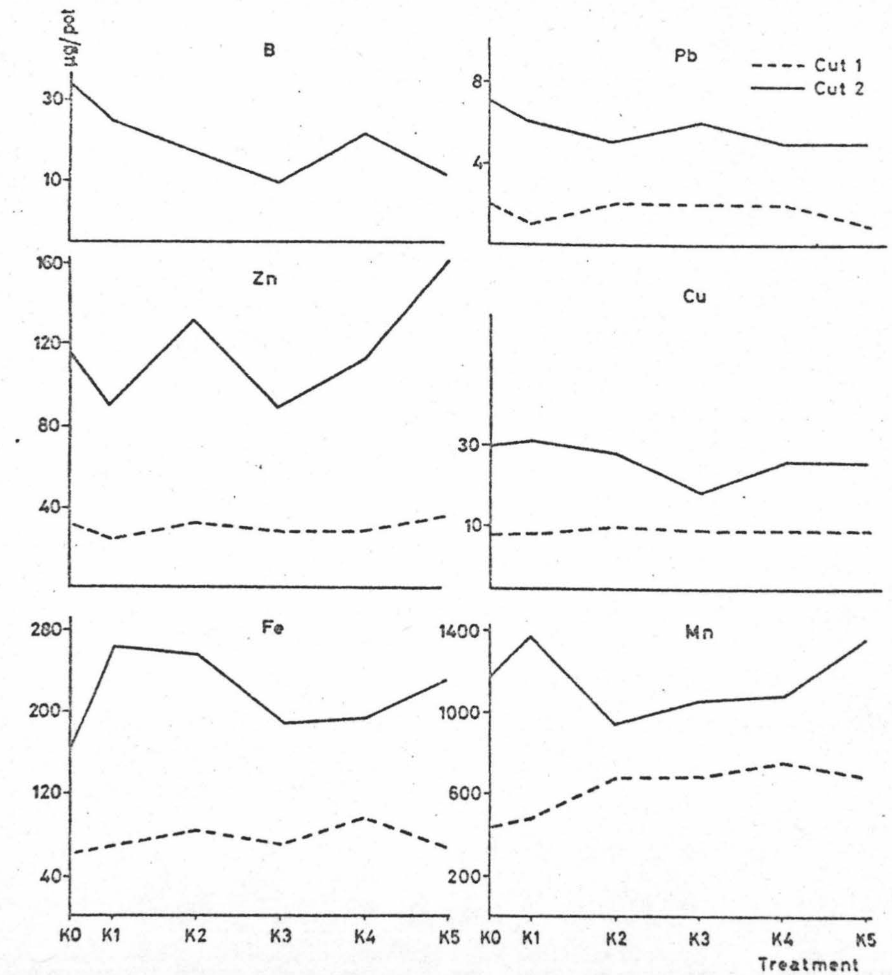


Table 4 : Statistical evaluation of trace element concentration (p.p.m. in dry matter) and uptake (μg/pot) as affected by addition of potassium sulphate

		Concentration						Uptake					
Regression	Cut	Fe	Mn	Zn	Cu	B	Pb	Fe	Mn	Zn	Cu	B	Pb
Linear	1	N.S.	**	N.S.	*	**	**	N.S.	**	**	N.S.	**	*
	2	**	**	N.S.	**	**	**	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
	Total	**	N.S.	N.S.	**	**	**	N.S.	**	*	N.S.	*	N.S.
Quadratic	1	N.S.	**	N.S.	N.S.	N.S.	N.S.	**	**	N.S.	N.S.	N.S.	N.S.
	2	N.S.	*	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
	Total	N.S.	N.S.	*	**	**	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
Cubic	1	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
	2	*	N.S.	N.S.	N.S.	*	N.S.	*	N.S.	N.S.	N.S.	*	N.S.
	Total	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	*	N.S.

* significant at 5 % ; ** significant at 1 % ; N.S. not significant

The pH was measured 14 days after the lime application giving the following changes :

Ca-level	pH-H_2O
Ca_O	6.30
Ca_1	6.65
Ca_2	7.15
Ca_3	7.30
Ca_4	7.45

3.2.4.1. Effect of Ca on the trace elements in the plants

Soil pH is a well known factor influencing the availability of trace elements, the general rule being that, except for Molybdenum, their availability decreases with increasing pH.
In this experiment the decrease in trace element content of the plant tissues, as a function of liming, was clearly observed for most of the elements under study, except for Fe, while also in the case of Cu the results were less regular (Fig. 8 & Table 5).
One of the elements known as giving the highest response towards pH variations is manganese (JONES, 1957 ; DE GROOT (1956) and RICH, 1956) ; however a wide divergence of opinious has been reported on the influence of Ca on the Mn nutrition of plants (CHAPMAN, 1931) ; SWANBACK, 1939), (MORRIS and PIERRE, 1947), TAPER and LEACH, 1957), (LOHNIS 196O) and others.
In our experiment the effect was markedly pronounced and the decrease in Mn content appeared progressively in function in increasing pH. The same progressive influence was observed for zinc, for which element analogous results were obtained by WEAR (1956).
In the cases of boron, copper and lead, the influence of liming was such that an important decrease occured already at the first Ca level. Therefore the controls were quite distinct from all the treatment levels. This results in a high frequency of significant quadratic regressions (Table 5).
Once again the antagonistic effect between Fe and Mn is appearing in some cases (SOMERS and SHIVE, 1942 ; MORRIS and PIERRE, 1947 ; SEDERIS and YOUNG, 1949). The scheme of our experiment

FIG. 8 INFLUENCE OF DIFFERENT LEVELS OF CALCIUM ON TRACE ELEMENT CONCENTRATIONS (P.P.m in dry matter) BY SHOOTS OF PERENNIAL RYEGRASS.

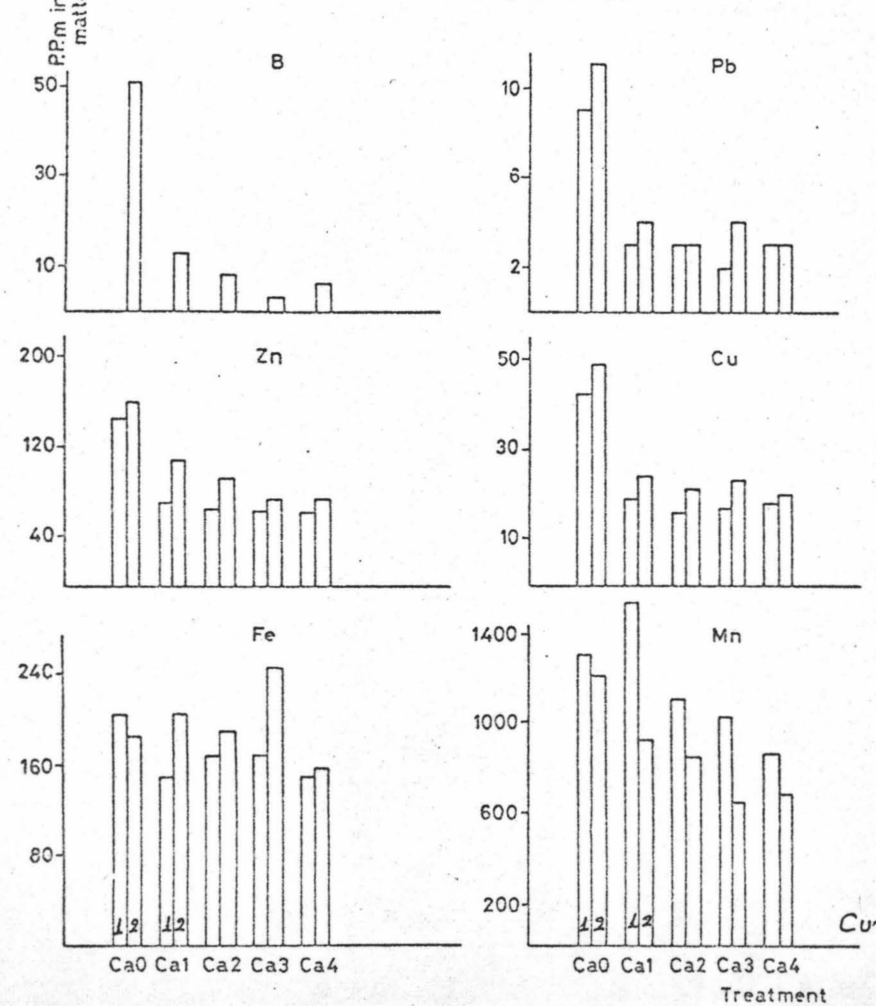


FIG. 9 INFLUENCE OF DIFFERENT LEVELS OF CALCIUM ON TRACE ELEMENT UPTAKE (μg/pot) BY SHOOTS OF PERENNIAL RYEGRASS

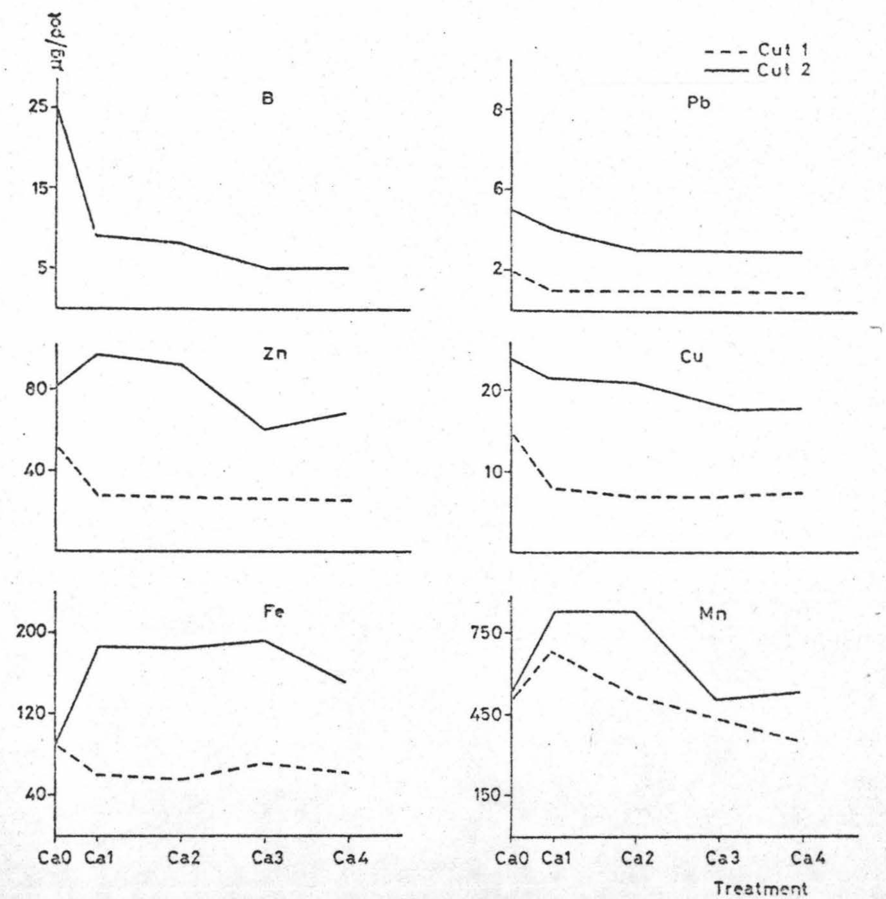


Table 5 : Statistical evaluation of trace element concentration (ppm in dry matter) and uptake (μg/pot) as affected by addition of calcium carbonate.

Regression	Cut	Concentration						Uptake					
		Fe	Mn	Zn	Cu	B	Pb	Fe	Mn	Zn	Cu	B	Pb
Linear	1	N.S.	*	**	**	tr.	N.S.	N.S.	N.S.	**	**	tr.	N.S.
	2	N.S.	N.S.	*	*	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
	Total	N.S.	*	**	**	–	*	**	N.S.	N.S.	N.S.	–	N.S.
Quadratic	1	*	**	**	**	tr.	*	N.S.	N.S.	**	**	tr.	N.S.
	2	N.S.	**	**	**	**	**	N.S.	*	N.S.	**	**	*
	Total	N.S.	**	**	**	–	**	N.S.	**	N.S.	**	–	**
Cubic	1	N.S.	N.S.	**	**	tr.	*	N.S.	N.S.	**	**	tr.	N.S.
	2	**	*	**	**	**	*	**	N.S.	N.S.	N.S.	N.S.	N.S.
	Total	N.S.	N.S.	**	**	–	**	**	N.S.	N.S.	N.S.	–	*

* significant at 5 % ; ** significant at 1 % ; N.S. not significant ; tr. traces

permits to confirm a calcium-Boron interaction (BERNNAN and SHIVE, 1948 ; GISIGER, 195O ; PURVIS et al., 1948 and MCILRATH and DE BRUYN, 1956).

The total uptake of trace elements as a function of liming has been largely influenced by the dry matter yield, which was itself influenced by the pH. The general trend is a decrease for most of the elements at higher Ca levels, but only in the cases of Cu and B this decrease started at the first liming level. Once again the significant regressions obtained were mainly of a quadratic order.

3.2.5. Effect of Magnesium

The influence of Mg on the concentration of different trace elements in the plants was rather limited. However, different significant regressions were observed (see table 6). The main point to be mentioned is a marked increase of Mn content between the Mg treated plants and the control in the first cut.

In the cases of Cu and Zn, the Mg treatment was acting in an opposite way and reduced the contents of these elements. The total uptake was generally the highest at the highest Mg level ; the two successive cuttings showed however an increase pattern for boron. The shape of the uptake curves was mainly influenced by the similar curves of the dry matter yields obtained.

4. GENERAL REMARKS AND CONCLUSIONS :

The experiments decribed here shows that even in a soil normally provided with nutrient elements, different interactions between major and trace element treatments could be observed. Indeed the yields in dry matter as well as the content and the total uptake of trace elements by Perennial ryegrass as an experimental crop, were in many cases affected by the applications of different levels of major elements in spite of the buffering effect of the original quantities already present in the soil at quite satisfactory levels. The major influence of the applied treatments started generally with the second cutting and the analysis of the results shows quite a number of significant and highly significant regressions.

FIG. 10 INFLUENCE OF DIFFERENT LEVELS OF MAGNESIUM ON TRACE ELEMENT CONCENTRATIONS (P.P.m in dry matter) BY SHOOTS OF PERENNIAL RYEGRASS

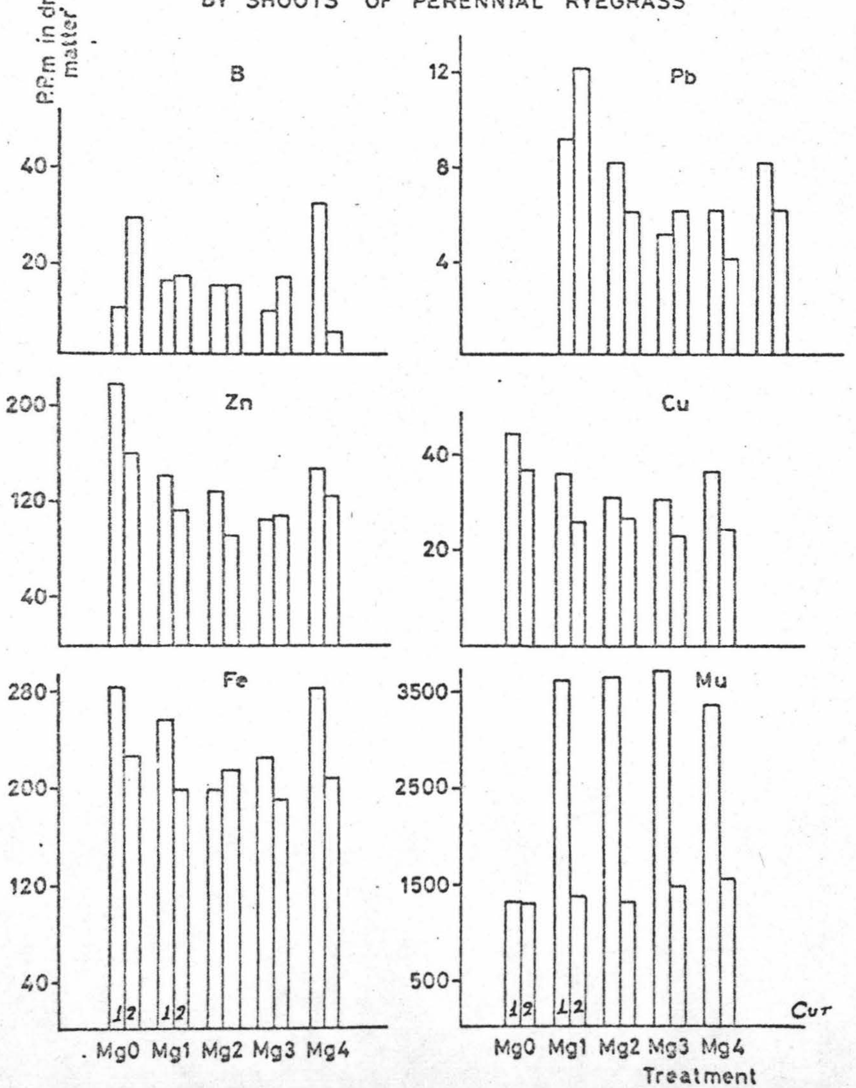


FIG. 11 INFLUENCE OF DIFFERENT LEVELS OF MAGNESIUM ON TRACE ELEMENT UPTAKE (μg/pot) BY SHOOTS OF PERENNIAL RYEGRASS

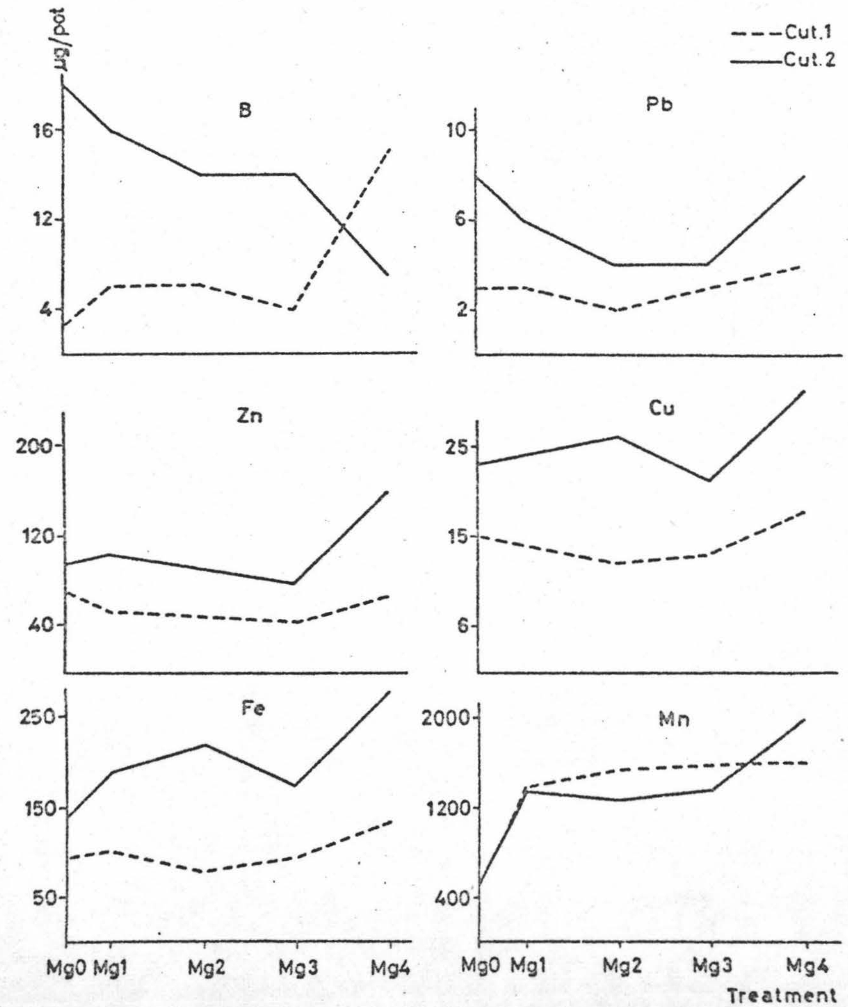


Table 6 : Statistical evaluation of trace element concentration (ppm in dry matter) and uptake (μg/pot) as affected by addition of magnesium sulphate.

		Concentration						Uptake					
Regression	Cut	Fe	Mn	Zn	Cu	B	Pb	Fe	Mn	Zn	Cu	B	Pb
Linear	1	N.S.	**	N.S.	N.S.	**	N.S.	**	**	N.S.	N.S.	**	N.S.
	2	N.S.	**	N.S.	**	**	**	**	**	**	**	**	N.S.
	Total	N.S.	**	N.S.	**	N.S.	N.S.	**	**	**	**	N.S.	N.S.
Quadratic	1	**	**	**	**	*	*	*	**	**	*	**	N.S.
	2	N.S.	N.S.	**	**	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	*
	Total	**	**	**	**	**	**	N.S.	**	**	*	N.S.	**
Cubic	1	N.S.	**	N.S.	N.S.	**	N.S.	N.S.	**	N.S.	N.S.	**	N.S.
	2	N.S.	N.S.	*	**	N.S.	**	**	**	N.S.	N.S.	N.S.	N.S.
	Total	N.S.	**	*	*	N.S.	N.S.	**	**	N.S.	N.S.	N.S.	N.S.

* significant at 5 % ; ** significant at 1 % ; N.S. not significant

Due to the fact that these appeared as cumulative functions of dry matter yield and concentrations of elements in the plant tissue, the regressions are often quadratic ones, that means curvilinear responses. The direct influence of the applied nutrient elements, was most pronounced in the cases of nitrogen and lime treatments and the elements boron, zinc and manganese were more affected than iron and copper. There was also a marked effect of K on the uptake of B and Pb.

CHAPTER III

THE EFFECT OF VARYING LEVELS OF TRACE ELEMENT FERTILIZATION ON TRACE ELEMENT UPTAKE BY GRASS GROWN UNDER GREENHOUSE CONDITIONS

1. INTRODUCTION

This work presents results of a pot experiment designed to determine the effect of various trace element application on grass production as well as on the levels of trace elementsin the plant tissue under greenhouse conditions.

The pot experiment was conducted in a greenhouse on three different soils. Fe, Mn, Zn,, Cu, B, Mo and Co were applied in varying levels with one dose of macronutrients including N, P, K and Mg.

Positive and negative responses of micronutrient applications towards the yields of different crops have been reported. MULVEHILL et al. (1955) report that boron, manganese, zinc and copper did not increase yields of lucerne or oats on 13 different soils, while yields of an lucerne-grass mixture were significantly increased by the addition of boron. TRUE and SHREWSBURG (1958) found molybdenum, copper and zinc, applied with superphosphate, to be effective in increasing the production of legumes on several Texas soils. Applications of these trace elements without superphosphate were ineffective. FINGER (1951) reports that boron depressed yeids of oats, clover and barley, while copper and manganese gave increased yields. SMELTZER et al. (1962) stated that herbage yields were not affected by cobalt, zinc, manganese, copper, sodium and molybdenum added in field trials on four different soils.

Similar trends of investigations were carried out by GRUHN et al. (1952) KLEINIG - LOVEDAY (1962), and KAG-KAGAS et al. (1964).

2. EXPERIMENTAL DETAILS

The present experiment was started in the first week of December 1968, on soils originating from :

A. Lemberge - sandy loam

B. Melle - sandy soil

C. Drongen - sandy soil

Before being retained for the present experiment these soils were analysed for pH, % C, C. E. C. and extractability of trace elements by nitric acid as an extractant.

Treatments :

Each soil was treated with different levels of trace elements mixture, and with a supplement of major elements. The trace elements were applied in solution ten days before sowing perennial ryegrass as mentioned in table 7.

Table 7 : Trace and macronutrients applied ten days before sowing Perennial ryegrass

	Trace elements ppm of element/ kg of soil			Major elements kg/ha	
Salt	1	2	3	Form	units
$MnSO_4 . 4H_2O$	2.5	5.-	10.-	NH_4NO_3	300
$CuSO_4 . 5H_2O$	2.5	5.-	10.-	$Ca(H_2PO_4)_2 . 2H_2O$	135
$ZnSO_4 . 7H_2O$	2.5	5.-	10.-	KCl	400
$Na_2MoO_4 . 2H_2O$	2.5	5.-	10.-	$MgSO_4$	100
$FeSO_4 . 7H_2O$	2.5	5.-	10.-		
$Na_2B_4O_7 . 10H_2O$	2.5	5.-	10.-		
$CoSO_4 . 7H_2O$					

Sixteen pots representing each soil, were divided into four treatments : control, 2.5 ppm, 5.0 ppm and 10.0 ppm of each trace element mixture, (table 7) replicated four times.

After mixing and potting, the pots were allowed to equilibrate for ten days before sowing 75 seeds of Lolium perenne L., perennial

ryegrass pasture type R. v. P. in each pot containing 1 kg of soil. The layout was completely randomised and two cuttings from soil A were harvested after 41 and 6O days respectively. The plants of soils B and C provided only one cutting.
Soil moisture was kept constant throughout the whole experiment at field capacity by the supply of deionized water. *

2. 1. Soil characteristics are given in tables 8 and 8. 1

Table 8

Soil	pH H_2O	pH KCl	C. E. C. meq/100g	% C
Soil A	5. 70	4. 85	11. 2	2. 30
Soil B	4. 95	3. 95	4. 5	1. 67
Soil C	4. 65	3. 85	6. 5	1. 70

Table 8. 1

	Extractable amounts of the trace elements ppm in air dry samples								
	Al	Fe	Mn	Zn	Cu	Pb	Mo	Co	Ni
Soil A O. 1 N HNO_3	265	175	40. 0	15	4. 0	8	2. 0	11	traces
O. 5 N HNO_3	410	760	140. 0	22	6. 0	4O	4. 5	16	2. 0
Soil B O. 1 N HNO_3	275	100	7. 5	10	1. 0	5	traces	10	3. 0
O. 5 N HNO_3	465	265	7. 5	31	13. 0	15	8. 0	12	7. 0
Soil C O. 1 N HNO_3	40	125	15. 0	15	4. 0	8	2. 0	11	3. 0
O. 5 N HNO_3	650	600	33. 0	15	8. 0	17	4. 0	11	3. 0

2. 2. Plant growth : The first cut was prolonged till 41 days in order to observe the behaviour of the plants under the influence of the different trace element levels. The growth period of the second cut was prolonged till 60 days because of the necessity of harvesting sufficient material for analysis. The second growth in soil A was very slow, while there was no more growth on soils B and C.

* Greenhouse temperature was + 15 °C and the plants were given artificial fluorescent light for 16 hours per day.

2. 3. Chemical analysis : At harvest the plants were cut above the soil level, dried at 70°C for 48 hours, and weight for dry matter yield (table 9). After ashing at 450°C for 4 hours, the ash was analysed for trace elements by direct reading spectrography (COTTENIE and GABRIELS, 1965). Soil extracts were analysed for trace elements by the same method.

Table 9 : Dry matter yields and ash percentage of perennial ryegrass grown on three different soils as influenced by different levels of trace element mixture.

		Soil A		Soil B		Soil C	
Trace element doses mg/kg soil	cut	Dry matter g/pot	% ash	Dry matter g/pot	% ash	Dry matter g/pot	% ash
O ppm	1	0. 396	19. 8	0. 349	19. 4	0. 437	19. 7
O ppm	2	0. 617	19. 0	0. 441	21. 8	0. 764	18. 1
2. 5 ppm	1	0. 530	22. 1	0. 196	21. 3	0. 312	19. 7
2. 5 ppm	2	0. 614	18. 7	-	-	0. 381	21. 7
5 ppm	1	0. 501	19. 1	0. 047	21. 3	0. 160	22. 0
5 ppm	2	0. 787	18. 7	-	-	-	-
1O ppm	1	0. 460	19. 3	0. 036	19. 9	0. 056	19. 3
1O ppm	2	0. 508	16. 1	-	-	-	-

3. RESULTS AND DISCUSSION

3. 1. Trace element caracterisation of the soils

Extractable values of Al, Fe, Mn, Zn, Cu, Pb, Mo, Co and Ni

The extractable values obtained (table 8. 1) of the above mentioned elements show that there was no indication of excess or deficiency in the three soils A, B and C.

Comparative mean figures of numerous samples analysed for the particular area in which the soils (A, B and C) were collected, are also reported (Dept. Report 1967 - 1969 - unpublished).

3.2. Dry matter yields as affected by different levels of trace elements applied on three different soils

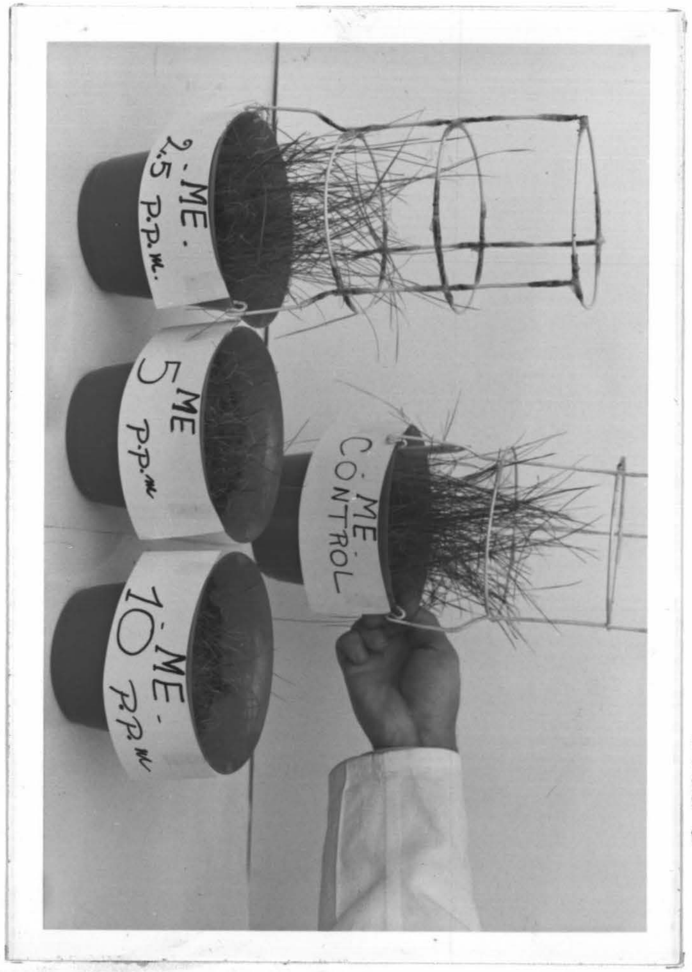
The effect of different levels of trace elements on dry matter yields of perennial ryegrass is tabulated in table 9. It has already been mentioned that only soil A (sandy loam) gave a second cutting. This was also the case for the first two levels of trace elements on soil C and for the control of soil B. From the present results it is evident that the trace element mixture strongly depressed the yields of perennial ryegrass on sandy soils, but had not the same pronounced effect on the heavier soil A (table 9). This is illustrated by the photographs given in Fig. 12. It seems that the effect of macronutrients on plant growth was more pronounced during the second growth period.

Since a marked decrease in dry matter yields corresponds to increasing rates of trace elements nutrition from plants grown on B and C soils, different significant regressions in plant uptake of Fe, Mn, Zn, Cu, B, Mo, Ni and Pb was recorded (tables 11 & 12).

3.3. Effect of trace element application of the iron content and uptake

In spite of the iron present in the trace element mixture a decrease of iron content in the plant shoots, grown on soil A, occured, except for the highest rate (10.0 ppm/kg soil) which caused an increase of iron content from 129 to 155 ppm. It seems that the soluble Fe applied has precipitated in the soil, while several other metal ions such as Cu, Zn, Mn, Mo and Ni may have affected the uptake of Fe ions. The antagonistic effect between Fe and other elements was already mentioned by SOMERS and SHIVE (1942) who reported that high concentrations of soluble (active) manganese in the plant tissues were invariably associated with low Fe concentrations. Iron uptake by plants is not only affected by pH and manganese but also by the levels of other elements, particularly phosphorus and heavy metals. DE KOCK and HALE (1955) found that the P/Fe ratio was higher in chlorotic leaves than in green ones. FORSTER (1954) observed heavy metal toxicities in soil, and DE KOCK (1954) in solutions and both found that one effect of these was an induced iron deficiency.

Soil C



Soil B

Soil A

Fig. 12 : Photographs illustrating the effect of trace element treatments applied to the three different soils, with perennial ryegrass used as a test crop.

The values of Fe content obtained on soils B and C were significantly higher than those on soil A. The treatments with 5.0 ppm soluble Fe in the mixture, gave 490 ppm and 235 ppm respectively. On soils B and C the Fe concentration of the plants increased quite linearly with increasing rates of Fe application, except for plants on soil B at the 10 ppm Fe treatment. A decrease from 490 to 320 ppm was recorded.

The Fe uptake of both cuts tends to decrease with the added trace element mixture.

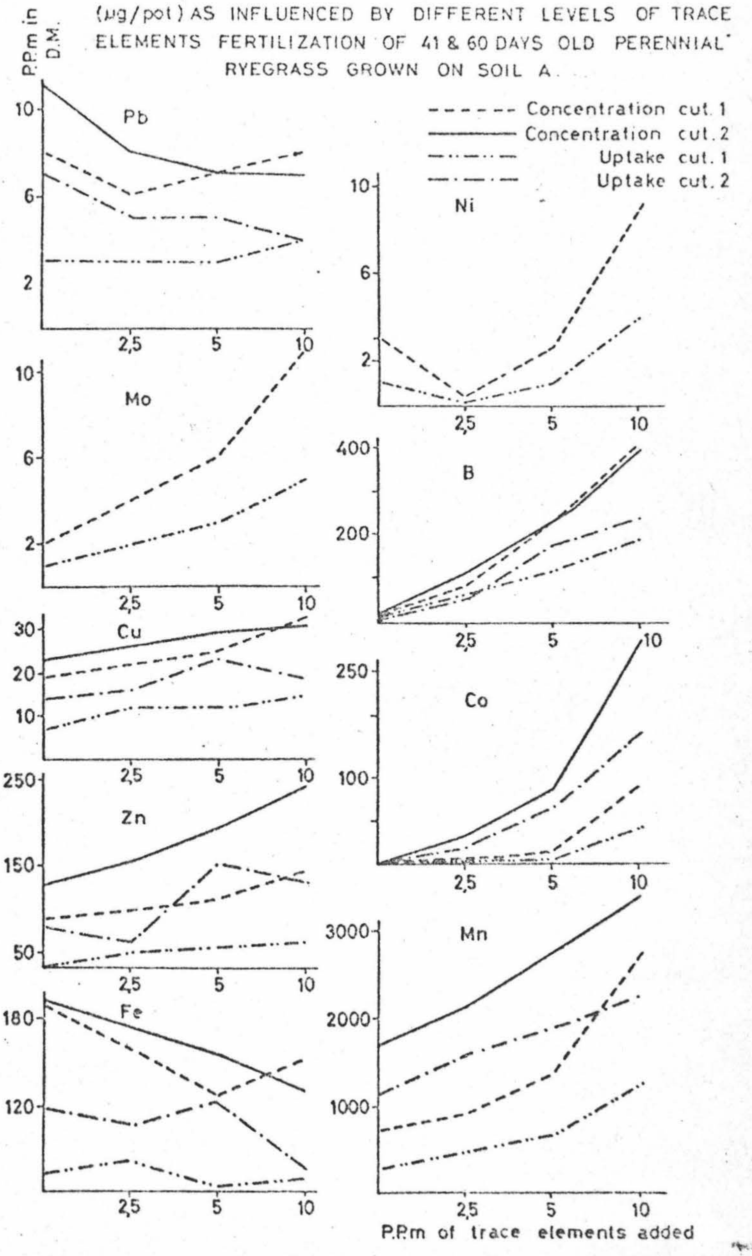
3.4. Effect of trace element applications on the manganese content and uptake

The manganese content of the plants grown on soil A, significantly increased with increasing rates of the trace element mixture (fig. 13). Other factors however, appear to have a secondary effect on the concentration of Mn in the plants. As indicated in table 11, high manganese concentrations (soil A), which may have reached toxicity, were obtained after application of 10 ppm Mn in the present mixture. Both cuts however followed a similar pattern, and this to emphasize the complete reverse pattern being indicated by the Fe concentration of the plants.
Manganese uptake followed a similar pattern as the content and a linear increase towards addition of Mn rates was observed (fig. 13). The treatment with 10 ppm soluble Mn in the trace element mixture produced plants with a different Mn status on the different soils : plants from A, B and C contained in the first cut 2749 ppm, 1510 ppm and 1540 ppm Mn respectively, and this in spite of soil A being less acid. Thus it appears that soluble Mn as well as pH of the soil, were the main factors deciding the enhancement of Mn content in the plant tissue. RICH (1956) and DE GROOT (1956) were in agreement with this.

3.5. Effect of trace element applications on the zinc content and uptake

The trace element mixture containing 2.5 and 5.0 ppm soluble zinc increased significantly the zinc concentration in the plant tissues in both cuts on soil A. This effect however was more pronounced in

FIG. 13 TRACE ELEMENTS CONCENTRATION (P.P.m in D.M.) AND UPTAKE (µg/pot) AS INFLUENCED BY DIFFERENT LEVELS OF TRACE ELEMENTS FERTILIZATION OF 41 & 60 DAYS OLD PERENNIAL RYEGRASS GROWN ON SOIL A.



the second cut. Since no dilution effect was observed, it appears clearly that the linear increase in zinc was mainly due to the zinc levels applied in the present mixture.
Linear increase in zinc content was similarly observed in the plants grown on B and C soils. These values were significantly higher than the ones obtained from plants of soil A (comparison based on first cut values). From the literature similar results were reported by different authors, KARLSSON (1952) and SHAW et al. (1954).
The uptake of Zn by the plants grown on Soil A, showed variations of response during the second cut ; the first cut was lineary significant in accordance to Zn applied.
On the other hand, a consistent decrease in zinc uptake from plants grown on soils B and C was obtained. These results however were not correlated with the high values of zinc concentration already discussed. This is due to the decrease in yields of dry matter in relation to the increase in trace element application.

3.6. Effect of trace element application on the copper content and uptake

The soluble copper applied in the trace element mixture, significantly increased the copper content of the plants grown on soils A, B and C. Higher values of copper however were observed in plants grown on B and C soils than on A soil. This may be due to the entry of copper in different reactions in the sandy loam soil A, which has the highest pH and the highest humus content, so that the availability of Cu was reduced. It has been pointed out that for cereals, the yields can increase by over 50 percent with copper application in copper deficient soils, without increasing the copper content of grain or straw (REITH and MITCHELL 1962-1964).
In the present experiment, as it was mentioned earlier, a sharp decrease in yield occured on soils B and C in function of the trace applications. MACKAY et al. (1966) and CHESTIRE et al. (1967) observed an increase of Cu content in the plant with fertilization on copper deficient organic soils. Our results with perennial ryegrass also show a marked increase of Cu content on soils B and C.

As the yields on these soils were significantly decreasing with trace application, the total uptake of copper however was also negatively influenced (fig. 14 & 15) while on soil A, in spite of a smaller change in content, there was till an increase of the total uptake.

Various opinions were reported on the relationship between soil pH and copper content WEHRMANN (1955), PIPER and BECKWITH (1951), PACK et al. (1953).

In spite of sometimes contradictory statements, our results indicate a strong dependence of the Cu-status of the plant towards soil characteristics and pH.

3.7. Effect of trace element application of boron content and uptake

Applications of boron significantly increased the boron contents of plants grown on soils A, B and C. However, the values obtained from plants on sandy loam (soil A) were considerably higher than those obtained from the sandy soils (B and C).

Both cuts show similar values of content in relation to the rates of boron applied.

On soil A the uptake as well as the content followed a linear increase (table 11). On soils B and C the content reached rapidly a maximum value of $\pm$ 250 ppm.

ANDERSON (1952) obtained good responses from lucerne and subterranean clover to 3.5 lb of borax per acre, and residual effects of this element were still evident after six years. On the other hand, MAC GREGOR and MULVEHILL (1955) found that 20 or 30 lb per acre of borax gave a significant increase in the boron content of lucerne and oats in the year of application only. The problem of the "residual effect" of boron, will be discussed in chapter V.

Large differences between the reaction of different grass species to excessive supplies of thi s element were reported by OERTLI et al. (1961) and OERTLI et al. (1961). Tables 10 & 10.1 point out the different values of boron content obtained under different growth conditions and equal amounts of boron supply. Table 10 was reported by OERTLI et al. (1961) and table 10.1 gives the results of the present experiment.

FIG 14 TRACE ELEMENTS CONCENTRATION (P.P.m in D.M.) AND UPTAKE (µg/pot) AS INFLUENCED BY DIFFERENT LEVELS OF TRACE ELEMENTS FERTILIZATION OF 41 & 60 DAYS OLD PERENNIAL RYEGRASS GROWN ON SOIL B.

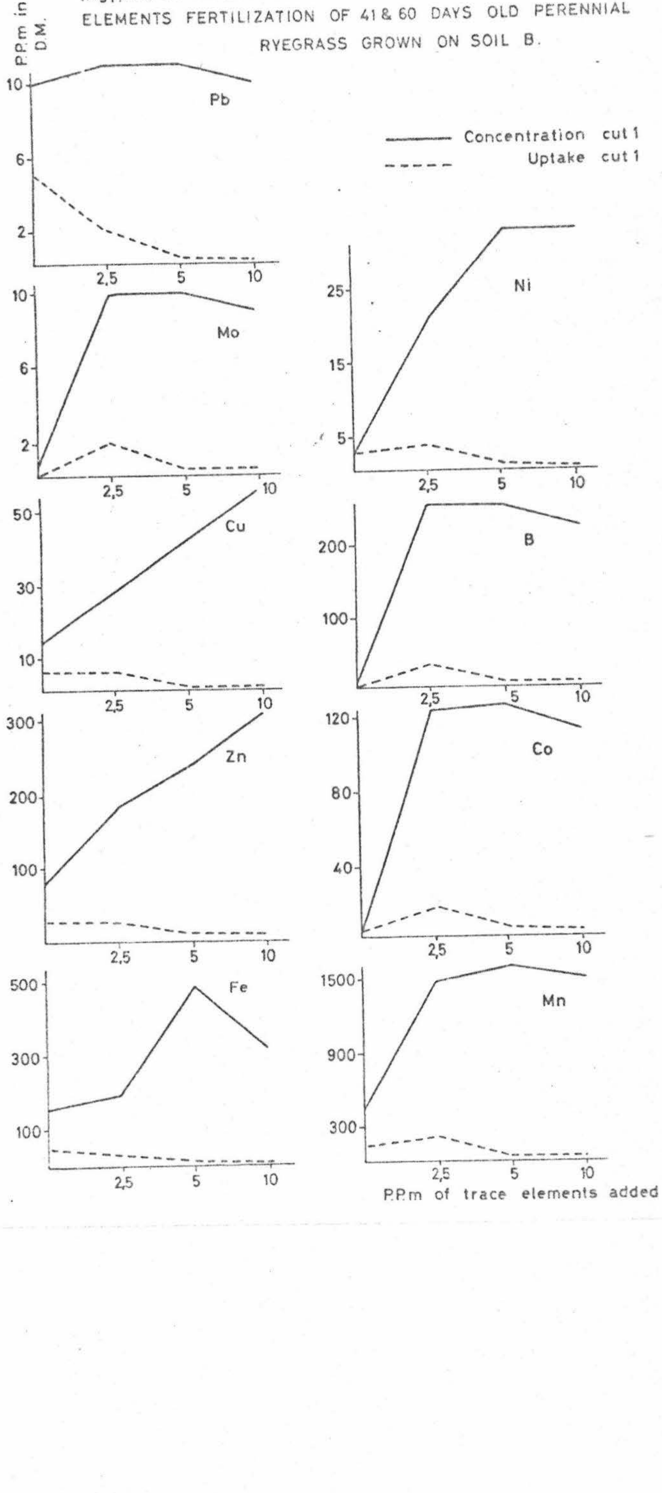


FIG. 15 TRACE ELEMENTS CONCENTRATION (PPm in D.M.) AND UPTAKE (ug/pot) AS INFLUENCED BY DIFFERENT LEVELS OF TRACE ELEMENTS FERTILIZATION OF 41 & 60 DAYS OLD PERENNIAL RYEGRASS GROWN ON SOIL C.

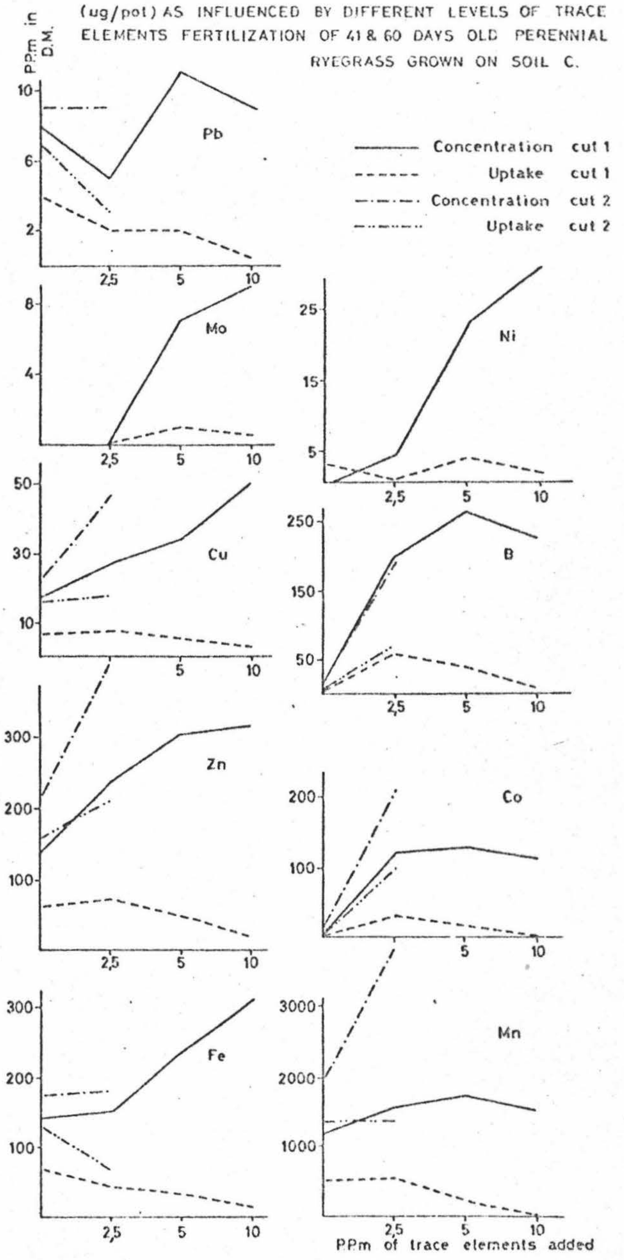


Table 10 : Time required to produce boron toxicity symptoms, and boron contents of leaves as given by OERTLI et al. (1961)

Species	Time required with 10 ppm B solution (days)	Boron content of leaves	
		Necrotic (ppm)	Green (ppm)
Alta fescue	8	1510 - 8200	50 - 760
Bluegrass	12	1860 - 6800	40 - 960
Per. ryegrass	8	950 - 2690	100 - 290
Bermuda grass	16	1380 - 5770	40 - 700

Table 10. 1 : Boron content of Perennial ryegrass shoots as affected by boron supply (10 ppm) after 41 days.

Soil-pH. H_2O		Boron content of leaves (ppm)
A	5. 7	412
B	4. 95	226
C	4. 65	231

In the more acid soils (B & C) the application of the first boron level (2. 5 ppm) already gave an important boron accumulation in the plants. The toxicity observed at higher levels of treatment, on these soils was no more corresponding with a further enrichment in the plant tissues. Therefore the total uptake in toxicity conditions was not higher than in normal conditions on these soils (fig. 14 & 15).

3. 8. Effect of trace element application on molybdenum content and uptake

The presence of molybdenum in the trace element mixture caused an important increase of the concentration of this element in the plant tissues, in comparison with their normal content. On each soil plants were harvested with molybdenum contents up to 9 or 10 ppm, but this was observed at the 2. 5 ppm level of treatment on soil B and at the 10 ppm level on soils A & C.
It is known that the pH of the soil is an important factor in deter-

mining the molybdenum availability, an increase of pH normally being in favour of the availability DAVIES et al. (1951) ; DAVIES (1965) ; EVANS et al. (1951) ; MC LACHLAN (1955) ; ROBINSON et al. (1951) ROBINSON and EDGINTON (1954) ; ROSSITER (1952) ; STEPHENS and OERTEL (1943) ; OERTEL et al. (1946) ; PIPER and BECKWITH (1951) ; STOUT et al. (1951) ; WALKER et al. (1955) ; DAVIES (1956).
In our experiment the highest molybdenum content in Perennial rye-grass (11 ppm) was also obtained on soil A, showing the highest pH. Other elements such as Ca, Mn and eventually sulphates may act as antagonists, but the experiment was not set up in order to observe such effects. The total uptake was only increasing with the application rate on soil A, but once again the uptake pattern on soils B & C was mainly influenced by the dry matter production.
It seems probably that the quantities of molybdenum applied may have influenced also the uptake of other ions, such as nickel, which was not present in the trace element mixture. This possibility was also suggested by OERTEL et al. (1946). As this experiment did not contain separate treatments, the effect of such interaction could not be further observed.

3.9. Effect of trace element application on cobalt content and uptake

Normally the cobalt content of pasture crops is found to be very small and values reported by HILL (1953) and MITCHELL (1954-1955) were not exceeding O.5 ppm.
SCHARRER and TAUBEL (1954) observed that a complete N-P-K fertilization resulted in a higher cobalt uptake. Concerning the relationships between the cobalt concentrations in soils and plants, different statements have been advanced. MOMCILO et al. (1961) stated that there is no direct relationship while WEHRMANN (1955) found the cobalt content of herbage to vary directly with the soil content though it was also affected by soil pH with which it varied inversely. While the original soil content was so low that it was impossible to observe any direct relationship between uptake and native content in the soil, the cobalt content of the plants increased significantly with increasing rates of cobalt application.

The increase was extremely high in comparison to the normal content of plants and reached levels of more than one hundred ppm, therefore it seems that this element may have been one of the main factors of the toxicity phenomenon observed at the higher levels of application. The slope of the curves showing the concentration of cobalt in the plant tissues in function of the treatments was once again less steep on soil A with the highest pH, but it reached the highest value (260 ppm) on this particular soil. On soils B & C the toxicity was thus, that no second cutting could be obtained. During the second cut on soil A the plant contents were much lower, showing an exortion and fixation of this element.

3. 10. Effect of trace element application of nickel and lead content and uptake

The present experiment permits also to make some observations concerning the uptake of nickel and lead, although these elements were not present in the trace element mixture. Therefore the differences which were noted must result from element interactions. CROOKE et al. (1954) observed that the nickel content of plants is reduced by high concentrations of iron in the substrate. A decrease of the ratio Ni/Fe in the growth medium decreased the uptake of nickel and thereby the intensity of its toxicity (CROOKE 1955). Both elements under study are indeed known as being toxic when their uptake is favoured.

Our analysis show that the nickel and lead uptake on soils B & C were more influenced by the treatments than on soil A, where the plant contents remained generally lower. In the case of nickel the plant contents strongly increased with the application of increasing amounts of the other trace elements, which is in contradiction to the observation of CROOKE. In the case of lead the influence was less pronounced and the values obtained on the three soils were quite similar. The range of lead content was indeed comparable to the one obtained in normal field conditions.

SUMMARY AND CONCLUSIONS :

Significant responses of Mn, Zn, Cu, B, Co and Mo concentrations in the plants as well as uptake with relation to the trace element applications were recorded (tables 11 & 12, figures 13, 14 & 15). The accumulation of higher values of some trace elements in the plant tissues in function of the rate of soluble elements added, was also found to be in relation to different soil characteristics. In general, the availability of these elements supplied to the soil is mainly related to the pH but other factors and interactions were also acting. Except on the sandy-loam soil with pH-H_2O 5.7 the perennial ryegrass suffered visibly from severe toxicity after the trace element treatments and in some cases the growth stopped completely after the first cutting. These toxicity phenomenon went together with highly increased contents of most of the trace elements in the plant tissues. The latter increase however, was somewhat levelled off in the soil with the highest pH for Fe and Cu, while also certain antagonisms were acting. The interactions between Fe and Mn as well as Fe and Ni could be confirmed by the results mentioned in this chapter. On the other hand the total uptake of trace elements was following a quite different pattern, due to the influence of the treatments on the growth and yields of the plants. The experiments support the point of view that the trace element content of ryegrass furnishes a quite true image of their availability in the soil. It is also an indication of quality of the grass production.

Table 11 : Trace elements uptake (μg) of 41, and 60 days old perennial ryegrass as influenced by different levels of trace elements applied to soils A and B.

			Uptake in μg per pot								
	Treatment	Cut days	Fe	Mn	Zn	Cu	B	Co	Ni	Mo	Pb
Soil A	Control	41	73.8	282.7	35.8	7.3	7.6	0.6	1.1	0.8	3.2
		60	120.3	1040.9	81.7	14.3	5.6	0.0	-	-	6.8
	2.5 ppm	41	84.8	497.3	52.4	11.8	57.2	1.8	0.2	2.1	3.3
		60	108.3	1281.2	65.5	16.3	48.2	15.8	-	-	5.1
	5.0 ppm	41	64.5	677.5	57.0	12.3	116.3	8.3	1.2	2.9	3.4
		60	123.5	2087.0	154.6	23.0	183.8	68.4	-	-	5.4
	10 ppm	41	71.3	1264.8	66.2	15.3	188.2	43.8	3.9	4.9	3.8
		60	79.2	2317.0	135.2	18.5	241.4	155.5	-	-	4.0
	Linear regression	41	N.S.	**	**	**	**	**	*	**	N.S.
	Quadratic "	41	N.S.	*	*	N.S.	N.S.	**	N.S.	N.S.	N.S.
	Linear regression	60	*	**	N.S.	**	**	**	-	-	N.S.
	Quadratic "	60	N.S.	N.S.	N.S.	*	N.S.	N.S.	-	-	N.S.
Soil B	Control	41	72.6	196.6	38.4	6.30	2.64	3.05	3.22	0.04	4.70
	2.5 ppm	41	35.7	214.3	36.4	5.70	48.72	23.42	3.69	0.92	2.10
	5.0 ppm	41	16.0	81.7	10.8	1.87	12.24	6.10	1.41	0.25	0.49
	10 ppm	41	11.3	53.3	10.0	1.97	7.99	4.00	1.18	0.33	0.35
	Linear regression		**	*	*	*	N.S.	N.S.	N.S.	N.S.	**
	Quadratic "		N.S.	N.S.	N.S.	N.S.	*	*	N.S.	*	N.S.

* significant at 5 % ; ** significant at 1 % ; N.S. not significant

Table 12 : Trace elements uptake (µg) of 41 days old perennial ryegrass as influenced by different levels of trace elements applied to soil C.

			Uptake µg per pot								
	Treatment	Cut days	Fe	Mn	Zn	Cu	B	Co	Ni	Mo	Pb
Soil C	Control	41	61.82	522.4	60.6	7.55	3.39	0.52	0.74	traces	2.59
	2.5 ppm	41	46.63	506.7	71.9	8.17	61.35	38.04	0.50	traces	1.73
	5.0 ppm	41	36.94	281.0	48.9	5.65	42.15	21.08	3.69	1.02	1.70
	10.0 ppm	41	17.20	83.3	17.4	2.75	13.80	6.40	1.69	0.47	0.52
	Linear regression		**	**	**	**	N.S.	N.S.	*	-	*
	Quadratic "		N.S.	N.S.	*	N.S.	**	**	**	-	N.S.

CHAPTER IV

INFLUENCE OF FARM YARD MANURE (F.Y.M.) AND N-P-K FERTILIZATION, APPLIED IN SANDY SOIL AT DIFFERENT RATIOS, ON THE TRACE ELEMENT UPTAKE OF PERENNIAL RYEGRASS UNDER GREENHOUSE CONDITIONS

1. INTRODUCTION

This pot experiment is dealing with the application of farm yard manure as an organic fertilizer to a sandy soil. In order to level off the differences in N, P, K, these elements were supplied in calculated amounts.

2. EXPERIMENTAL DETAILS :

This pot experiment was conducted in march 1968, on a sandy soil collected from an uncultivated area, located near the experimental farm of the Agricultural Faculty (Melle). The chemical characteristics of this soil were determined as follows :

pH-H_2O	4.85
pH-KCl	3.95
C.E.C.	4.5 meq/100 g

Before sowing and after the last harvest (3rd) the soil was analysed for the trace elements as extracted by nitric acid (table 13).

Table 13 : Trace elements in soil (extracted with O.5 n HNO_3)

	g FYM kg of soil	Fe	mg/kg of soil Mn	Al	Zn	Cu	Mo	Co	Ni	Pb
pre-sowing	0	4950	60	4400	20	2	7	10	33	65
After the 3rd cut	35	2200	60	3600	60	4	25	10	tr.	12.5
After the 3rd cut	70	2800	60	3600	60	4	52.5	25	15	15

Before being retained for the present experiment, the farm yard manure was analysed and the results are given in table 13. 1.

Table 13. 1. : Extractable amounts of the trace elements in F. Y. M.

	ppm in air dry sample								
	Fe	Mn	Al	Zn	Cu	Mo	Co	Ni	Pb
O. 1 N HNO_3	17. 5	44. 0	7. 5	28. 0	0.70	tr.	tr.	tr.	3. 0
O. 5 N HNO_3	73. 0	84. 0	27. 0	53. 0	3. 25	tr.	tr.	tr.	6. 0

The soil was treated with different rates of F. Y. M. as shown in table 14.

Table 14 : Quantities of F. Y. M. applied

Treatment No	quantity of sandy soil in grams	quantity of F. Y. M. in grams	correspon-ding quanti-ty of F. Y. M. in kg/ha °
1	1. 000	0	0
2	995. 5	4. 5	6. 250
3	991. 0	9. 0	12. 500
4	982. 5	17. 5	25. 000
5	965. 0	35. 0	50. 000
6	930. 0	70. 0	100. 000

The corresponding quantities of F. Y. M. per ha were calculated as shown by the following example :

1 kg contains 17. 5 g F. YM. = 0. 0175 kg F. Y. M.

1. 425. 000 kg contains 0. 0175 x 1. 425. 000 kg F. Y. M. = 24. 927 kg F. Y. M.

1 ha = 1. 000. 000 dm^2 x 0. 95 dm (pot depth) = 950. 000 dm^3

950. 000 x 1. 5 = 1. 425. 000 kg

° calculated on the basis of a depth of 9. 5 cm of soil, as in the pots.

2. 1. Treatments

In addition to the N-P-K introduced as farm yard manure, the different pots were also supplied with nitrogen as NH_4NO_3, phosphorus as $Ca(H_2PO_4)_2$ and potassium as K_2SO_4 in order to recieve finally the same amounts of each of these elements. The N-P-K values in the F. Y. M. were calculated on the basis given by ELSEVIER (1954), LANDBOUWGIDS (1957) and VERBELEN (1960). Table 15 shows the details of these treatments.

Table 15 : Quantities of N, P, K applied (in kg per ha)

	application as F. Y. M.			application of chemical fertilizers before sowing			application of chemical fertilizers after 1st cut			application of chemical fertilizers after 2nd cut		
F. Y. M g/kg soil	N	P_2O_5	K_2O	N	P_2O_5	K_2O	N	P_2O_5	K_2O	N	P_2O_5	K_2O
Control	-	-	-	-	-	-	-	-	-	-	-	-
4. 5	10	12. 5	31. 5	90	90	70	100	100	100	100	100	100
9. 0	20	25	63. 0	80	75	40	100	100	100	100	100	100
17. 5	40	50	125	60	50	-	100	100	100	100	100	100
35. 0	80	100	250	20	-	-	100	100	-	100	100	50
70. 0	160	200	500	-	-	-	40	-	-	100	100	-

2. 2. Procedure

The whole experiment consisted of 24 pots representing six different treatments with four replications : Control, 4. 5, 9. O, 17. 5, 35 and 7O g of F. Y. M.

After mixing the F. YM. + N-P-K with the soil and potting, the pots were allowed to equilibrate for four days more before sowing 75 seeds of Lolium perenne L. , perennial ryegrass (pasture type C. V. Vigor) in each pot containing 1 kg of soil. The layout was completely randomised, and three cuts were harvested with intervals of 21 days. The other factors such as soil moisture and artificial light were similar to the ones already mentioned in the previous pot experiment.

3. RESULTS AND DISCUSSION

3. 1. Dry matter yields as influenced by F. Y. M. and N-P-K fertilizers

Since the sandy soil was a poor source of major element nutrition, any level in the present experiment played an important role. The positive responses of dry matter yield towards the rates of farmyard manure is clearly illustrated by the histograms given in fig. 16 a.

In the first two cuts the maximum dry matter yields were reached after treatment with 35 g F. Y. M. /kg (for N-P-K see table 15). The most important differences in yields were obtained in the second cutting, and this shows that the F. Y. M. started to be really effective mainly after a certain period of its incorporation in the soil. The third cut seems not to follow the pattern observed in the first two cuts, a maximum yield of dry matter being recorded at the ratio of 17.5 g F. Y. M. per kg of soil, combined with N P K 100 : 100 : 100, supplied after the first and the second cuts respectively. Considering the total production of the three cuttings we may conclude that the yields were significantly influenced by the increasing levels of F. Y. M. The effect of F. Y. M. was also quite remarkeable on the root production but not systematic, as was the case with the shoots. The maximum root production corresponded with the treatments of 17.5 g of F. Y. M., while the other treatments had a similar effect on the root dry matter yields (fig. 16 b).

3. 2. Effect of F. Y. M. on the iron content and uptake

Very small variation of Fe concentration were observed between the different treatments.

The combination of the lowest rate of F. Y. M. with the chemical forms of N-P-K provided higher iron concentrations in the shoots, the iron content of the grass showing a tendency to decrease by the addition of F. Y. M. (table 21). In connection to this point MILLER and OHLOROGGE (1958) stated that chelating compounds present in manure held iron and zinc in forms less available to plants.

It is also noteworthy that the first cut produced in general plants with the highest iron concentration, followed by the third, while the second had the smallest iron values. This means that the

FIG.16a. THE EFFECT OF DIFFERENT LEVELS OF FARM YARD MANURE (F.Y.M.) AND N.P.K. FERTILIZERS ON THE DRY MATTER YIELDS (mg/kg soil) OF PERENNIAL RYEGRASS OVER THREE CULTINGS.

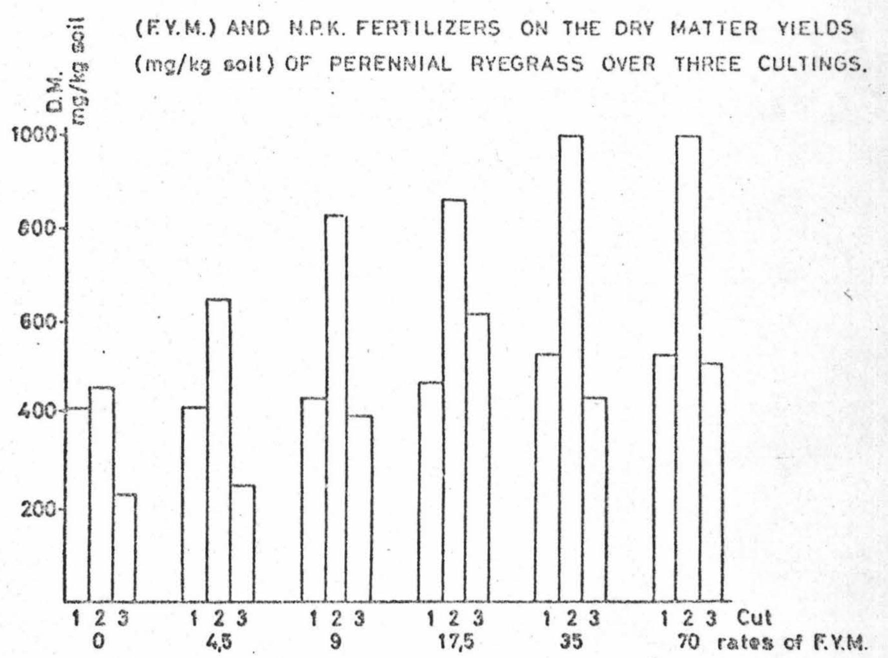
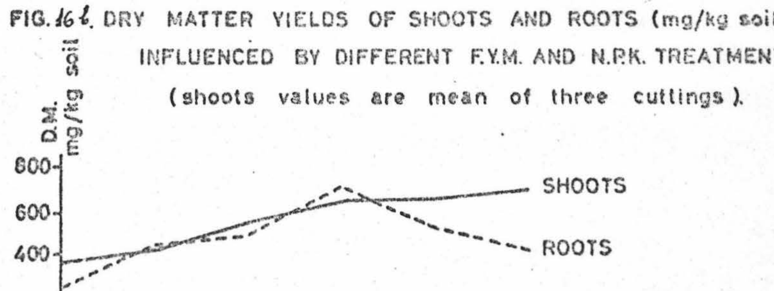


FIG.16b. DRY MATTER YIELDS OF SHOOTS AND ROOTS (mg/kg soil) AS INFLUENCED BY DIFFERENT F.Y.M. AND N.P.K. TREATMENTS. (shoots values are mean of three cuttings).



higher yields of the second cut were accompanied by a certain dilution effect.

Compared to the shoots the Fe concentration was more variable in the roots. The ratio of Fe in roots to tops was rather high, which indicates that the amount of Fe in the upper part depends not only on the rate of root accumulation (table 16).

The relationships of this element in the shoots and roots on the basis of content and uptake are given in table 16.

Table 16 : Iron content (p. p. m.) in dry matter and uptake (µg/pot) of roots of perennial ryegrass aged 63 days.

	g F. Y. M. per kg soil					
	Control	4. 5	9. 0	17. 5	35	70
Shoot content	213. 0	141. 6	129. 8	110. 9	124. 8	128. 6
Root content	2221	1439	1550	1173	2191	2014
Content ratio shoot/root	0. 10	0. 10	0. 08	0. 10	0. 06	0. 06
Shoot uptake	88. 42	60. 43	67. 35	69. 69	76. 25	85. 64
Root uptake	642. 6	690. 4	588. 6	849. 5	1217	890. 9
Uptake ratio shoot/root	0. 12	0. 09	0. 10	0. 08	0. 06	0. 11

In general, the total uptake of Fe was mainly governed by the total dry matter production.

3. 3. Effect of F. Y. M. on the manganese content and uptake

The fertilizer effect on the Mn concentration showed a steady increase of Mn content with decreasing F. Y. M. and increasing N-P-K fertilizer applications (table 21).

WILLIAMS (1960) stated that F. Y. M. and a complete N-P-K fertilizer had a similar effect on the proportions of Cu, Mn, Mo and Zn in wheat, barley, clover, potatoes and kale. MILLER and OHLROGGE (1958) concluded that manganese availability was increased by manure applications. Both these statements are not in agreement with the present data. The reason for this is probably the acidity of the soil being used in this experiment. Therefore, in spite of the fact that the soil was relatively low in Mn content, this element was present in an available form. This also explains the fact that high Mn concentrations were found in the control plants and in the ones with the lowest F.Y.M.-treatments. The application of 4.5 g F.Y.M./kg (lowest), combined with the highest N-P-K level gave more than three times the Mn values obtained at the rate of 70 g F.Y.M./kg, 2.5 times those with rate 35 g/kg and 2 times those with 17.5 g F.Y.M./kg.

Concerning the successive cuts, the second one, giving the highest yields, showed the maximum Mn values. The total uptake of Mn showed a completely analogous picture. This is in favour of the assumption that the N-P-K treatment more strongly influenced the Mn absorption than did the F.Y.M.

There were no significant differences between the Mn values accumulated in the roots to those of the shoots. The pattern was thus, that a continuous decrease of Mn content corresponds with the increase of F.Y.M. applications (except for the treatment with 35 g F.Y.M.). Unlike Fe, the translocation of Mn from root to tops seem to be less involved with other factors, and in some cases the shoots accumulated higher rates of manganese than the roots (table 17).

Table 17 : Influence of various levels of F.Y.M. on the manganese content (ppm in dry matter) and uptake μg/pot of Perennial ryegrass aged 63 days.

	g F.Y.M. per kg soil					
	Control	4.5	9.0	17.5	35	70
Shoot content	621.6	851.0	616.6	373.7	329.6	278.8
Root content	788.0	739.0	516.0	491.0	583.0	342.0
Content ratio shoot/root	0.79	1.15	1.09	0.76	0.57	0.82
Shoot uptake	221.8	369.7	342.8	260.2	236.0	197.0
Root uptake	265.3	344.2	285.8	350.1	300.5	152.8
Uptake ratio shoot/root	0.84	1.10	1.20	0.74	0.79	1.30

3.4. Effect of F. Y. M. on the zinc content and uptake

The effect of F. Y. M. combined with N-P-K, resulted in a negative response with regard to the Zn concentration. The higher rates of F. Y. M. with the lowest levels of N-P-K (i. e. 70 and 35 g of F. Y. M. /kg of soil) gave the lowest Zn concentrations in the shoots. The rates of 9 and 4.5 g F. Y. M. /kg of soil with the highest application of N-P-K. corresponded with the maximum of Zn contents. It appears that the highest Zn values correspond with the highest N-P-K treatments. It has been reported that an increase of Zn is mainly due to nitrogen application OZANNE (1955), GOMIDE et al. (1969) . This experiment confirms that the F. Y. M. had less effect on the Zn content of grass, compared with the Zn values obtained from the treatments recieving high levels of N-P-K fertilizer.

Concerning the total Zn uptake, the differences in content were levelled off by the differences in dry matter production (fig. 17), and any increase of Zn uptake went parallel with an increase of dry matter yield and vice versa.

There was a quite constant relationship between the zinc concentration of the roots to those of the shoots, the root concentration being meanly 5 times higher (table 18).

Thus, similar to Fe, quite high amounts of Zn were accumulated in the roots compared to the values found in the shoots. This may correspond with the observation of MILLER and OHLOROGGE (1958) who found that chelating compounds present in manure held Fe and Zn in forms less available to plants.

FIG. 17. THE EFFECT OF F.Y.M. AND N.P.K. FERTILIZERS ON THE CONCENTRATION (P.P.m in dry matter) AND UPTAKE (μg/pot) OF Fe, Mn, Zn, AND Cu. VALUES ARE MEANS OF THREE CUTTINGS

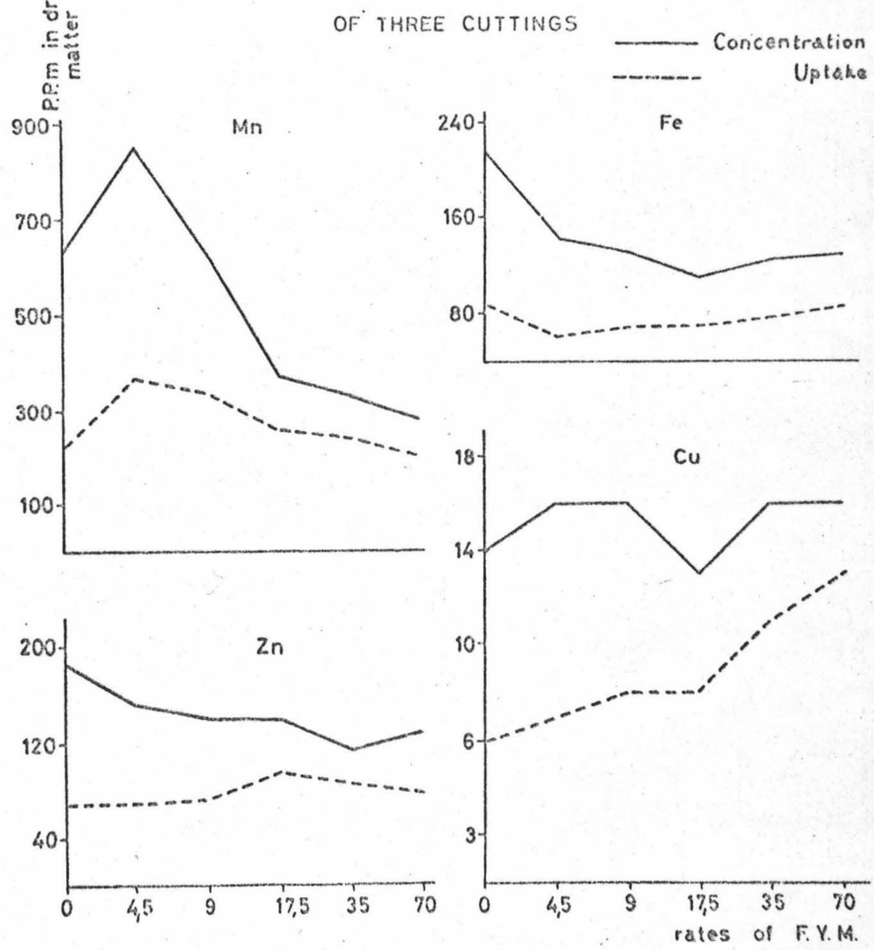


Table 18 : Influence of various levels of F. Y. M. and N-P-K on the zinc content (ppm in dry matter) and uptake µg/pot of perennial ryegrass aged 63 days.

		g F.Y.M. per kg soil				
	Control	4.5	9.0	17.5	35	70
Shoot content	178.2	151.2	140.8	140.9	114.3	127.4
Root content	647.0	763.0	662.0	672.0	672.0	730.0
Content ratio shoot/root	0.28	0.20	0.21	0.21	0.17	0.17
Shoot uptake	66.73	67.90	72.79	96.69	85.94	77.93
Root uptake	146.1	364.3	325.3	479.8	365.5	334.9
Uptake ratio shoot/root	0.46	0.19	0.22	0.20	0.24	0.23

3.5. Effect of F.Y.M. on the copper content and uptake

The Cu content was not significantly affected by both F. Y. M. and N-P-K. At the highest rate of F. Y. M. and the lowest dose of N-P-K similar Cu concentrations were found as the lowest rate of F. Y. M. and highest level of N-P-K. As a matter of fact, the Cu concentrations observed here were moderate which corresponds with the observation of WILLIAMS (1960) that N-P-K and F. Y. M. depressed the percentage of Cu, Mo, and Zn more often than they increased them.

The explanation given was that, due to the treatments, yields increase while the micronutrients are diluted in the harvested crop. In spite of this, the Cu concentrations obtained in the second cut, where the maximum yields were recorded, were not systematically less than those obtained in the other cuts.

The total Cu uptake followed a systematic increase with the F. Y. M. treatments. At the highest rate of F. Y. M. and the lowest level of N-P-K, Cu uptake reached the maximum. The results in table 19 show that the Cu content of the roots is systematically higher than in the shoots.

The roots as well as the shoots absorbed the lowest rates of Cu when only N-P-K fertilizers were applied (Control).

Table 19 : Influence of various levels of F. Y. M. and N-P-K on the copper content (ppm in dry matter) and uptake μg/pot of perennial rye grass aged 63 days.

	g F. YM. per kg soil					
	Control	4. 5	9. 0	17. 5	35	70
Shoot content	13. 95	15. 88	16. 37	13. 27	16. 79	15. 64
Root content	37. 00	31. 95	38. 90	38. 10	44. 42	54. 00
Content ratio shoot/root	0. 38	0. 50	0. 42	0. 35	0. 38	0. 29
Shoot uptake	5. 65	7. 32	7. 67	8. 36	11. 43	13. 29
Root uptake	10. 75	15. 42	19. 64	27. 32	24. 63	24. 42
Uptake ratio shoot / root	0. 53	0. 47	0. 45	0. 31	0. 46	0. 54

3. 6. Effect of F. Y. M. on the boron content and uptake

The rates of F. Y. M. and N-P-K fertilizers had no marked effect on the boron concentration in the grass.
In spite of quite large variations of the observed values, similarly to what has been observed in the cases of Mn and Zn, the highest concentrations were found in the second cut, which also produced the highest yields. Obviously there has no been any dilution effect observed.
The total absorption pattern of the successive cuts followed similar trends as the boron concentration.

Table 2O : Influence of various levels of F. Y. M. on the boron content (ppm in dry matter) and uptake µg/pot of perennial ryegrass aged 63 days.

	g F. Y. M. per kg soil					
	Control	4.5	9.0	17.5	35	70
Shoot content	30.00	35.62	36.53	29.34	24.13	39.61
Root content	7.68	33.40	48.10	44.80	45.40	54.90
Content ratio shoot/root	3.91	1.10	0.96	0.65	0.53	0.72
Shoot uptake	9.28	19.46	23.31	19.85	18.25	29.28
Root uptake	20.25	17.61	24.94	31.35	24.20	23.82
Uptake ratio shoot/root	0.46	1.11	0.93	0.63	0.75	1.23

4. SUMMARY AND CONCLUSIONS

The experimental study of the influence of F. Y. M. combined with N-P-K fertilizer on the behaviour of perennial ryegrass (pasture type R. v. P.) was observed during three successive cuts in a pot experiment.
The dry matter production was positively influenced by the treatments. The absorption of trace elements showed different patterns in function of the elements under investigation.
So for Fe and Cu the concentrations in the plant tissues were generally not significantly influenced, while significant regressions were observed for the elements Mn and Zn and to a certain extent for B. The highest dry matter production was obtained in the second cut, and this went together with the highest concentration for the elements Mn, Zn and B. This observation shows that the well known "dilution effect" is not to be considered as a general rule and that a considerable increase in dry matter production is not necessarily linked with lower mineral element contents. Moreover the organic fertilizer applied in the form of F. Y. M. appears to suppress the uptake as well the concentrations of Mn and Zn. The total uptake

of trace elements during the successive cuttings shows that for some elements the dry matter production was the most important factor, while for other elements the differences in concentrations were dominating.

Table 21 : Effect of F.Y.M. and N-P-K fertilisations on the trace element concentrations (p.p.m. in dry matter) of perennial ryegrass over three cuttings

g F.Y.M. per kg	Cut	ppm in dry matter Fe	Mn	Zn	Cu	B
0	1	257	530	182.4	23.7	37.2
	2	213	268	131.4	7.7	37.4
	3	168	1066	220.7	10.3	15.3
4.5	1	136	374	126.7	18.1	29.4
	2	131	1171	175.6	17.5	67.4
	3	157	1008	151.2	12.0	9.9
9	1	152	328	122.8	18.9	25.3
	2	111	867	159.3	17.7	76.2
	3	116	653	140.3	12.5	8.0
17.5	1	136	258	126.8	17.2	32.0
	2	91	484	180.0	11.8	47.2
	3	105	379	116.1	10.9	8.9
35	1	135	250	86.4	20.4	20.1
	2	98	371	144.4	17.4	39.8
	3	141	368	112.0	12.6	12.5
70	1	180	251	149.2	18.9	49.8
	2	110	314	120.5	22.8	50.6
	3	96	275	117.4	13.3	18.4
Quadratic regression	1	**	**	**	N.S.	**
	2	N.S.	N.S.	N.S.	N.S.	N.S.
	3	N.S.	**	**	N.S.	*
Quadr. reg.	mean total	N.S.	N.S.	N.S.	N.S.	N.S.
Lin. reg.		N.S.	**	*	N.S.	N.S.

CHAPTER V

A. EFFECT OF MICRO AND MACRONUTRIENTS FERTILIZERS ON PASTURE CROPS (FIELD EXPERIMENTS)

A. 1. LONG TERM FIELD EXPERIMENT (Since 1939)

1. INTRODUCTION :

In this chapter the results are discribed of the study which we had the opportunity to make concerning the trace element absorption by pasture crops growing on a long term experimental field. The different plots were subjected since 1939 to repeated one-sided treatments with major elements, while also trace element treatments were applied in 1967 and 1968.

In the present investigation an attempt was made to study under field conditions the following points :

1. The influence of long term one-sided fertilization on the trace element absorption by the pasture plants.
2. The effect of trace element fertilization
3. The variations in trace element absorption due to seasonal effect.
4. The botanical composition as a factor influencing mineral composition of the pasture crops.

2. EXPERIMENTAL DETAILS :

2. 1. Site and cropping history :

The permanent trials are situated in Melle and started in 1939, the field being divided into two main parts indicated as M. 39. 1. and M. 39. 2. The present study was conducted during two years of growth, extending from the beginning of 1967 to the end of 1968.

- M. 39. 1. : from 1939 till 1962, crops as grass (2 x), maize, potatoes, red clover, Italian ryegrass (2 x), lucerne, and tobacco were rotationaly grown till the year 1962.

  Present crops : in may 1963 the following mixture of species was sown :

kg/ha	2.250	Lolium perenne L., perennial ryegrass pasture-type C.V. 'Vigor'.
	2.0	Lolium perenne L., perennial ryegrass hay pasture-type C.V. 'Melino'.
	10.0	Festuca pratensis Huds., meadow fescue C.V. 'Merbeem'.
	2.2	Pleum pratense L., timothy, C.V. "Erecta R.v.P.".
	2.2	Dactylis glomerator L., cocksfoot, C.V. 'Lemba R.v.P.'.
	3.0	Festuca rubra L., red fescue.
	1.5	Poa trivialis L., rough stalked meadow grass
	1.0	Poa pratensis L., smooth stalked meadow grass C.V. 'Mervel'.
	3.0	Trifolium repens L., white clover C.V. 'Blanca R.v.P.'.
	0.5	Agrostis tenuis Sibth., browntop.

- M.39.2. : From 1939 till 1948 a mixture of grasses and clover was grown.

  Present crops : From 1948 till the present time, the grass and clover mixture sown was in kg per ha :

  Lolium perenne L. : 25

  Festuca pratensis Huds : 10

  Trifolium repens L. : 5

  Lolium multiflorum ; Italian ryegrass : 4

2.2. Fertilizer treatments

- Major elements : there were seven treatments with three replications as follows :

  1. Control
  2. N-P-KCa
  3. P-KCa
  4. N-KCa
  5. N-P-Ca
  6. N-P-K
  7. N-P-KCaMg

In 1967 a mixture of microelements were applied to M. 39. 1. a (2O kg/ha).

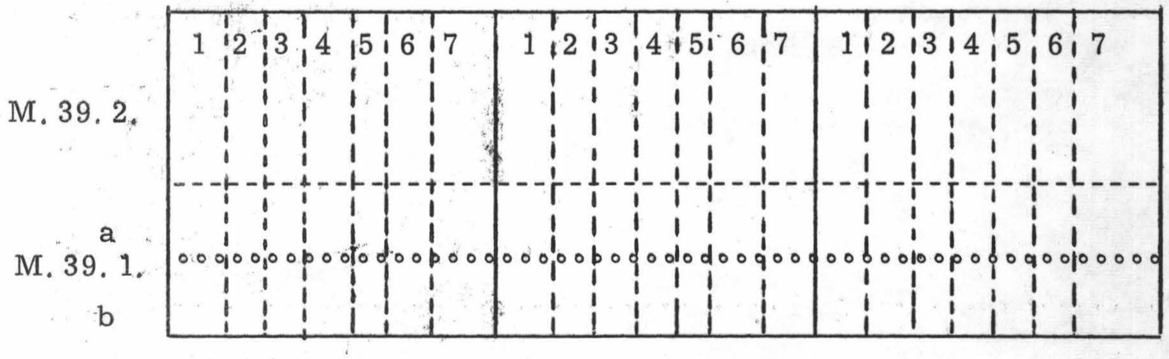


Table 22 : Amounts of fertilisers applied in kg/ha/year *

Years	N		P_2O_5		K_2O		CaO			MgO	
	dose	form	dose	form	dose	form	dose as $CaCO_3$	with NPK	sum	dose	form
From '39-'47	50	Am. sulfate	90	Super 18	120	Potassium-chloride 40	152	140	302	55	Mg-lime
From '48-'57	133	Am. nitrate 20.5	133	Super 18 or basic slags 18	150	" 40	212	344	556	76	Mg & Ca carbonate
From '58-'61	220	Am. nitrate 20.5 or 22.5	180	Super 18 or $CaHPO_4$ 38	160	" 40	67	437	504	23	Mg-carbonate
1962	315	Am. nitrate 22.5	150	Triple super 43	400	" 40	-	386	386	60	Mg-Ca carbonate
From '63-'65	307	Urea 45	145	Triple 43	400	" 40	117	70	187	30	Mg & Ca carbonate
From '66-'68	300	Am. nitrate 33	150	Triple 40	400	K-sulphate 50	177	80	257	100	Mg-sulphate

* Data obtained from the "Centrum voor Grasland onderzoek" - Melle (Dir. Prof. M. SLAATS).

2. 3. Trace elements

The M. 39. 1. plots, as indicated by the scheme, were divided into two parts during both years (1967 & 1968) of the study. On the first part, which is indicated by a, the plots recieved a mixture of trace elements, while the second part b received only the treatment with major elements. The trace element mixture (20 kg/ha) was applied as solution, sprayed on the foliage of the plants in the year 1967. The same dose of a mixture with different ratios was applied as a powder in two separate doses during 1968.

The powder was mixed with sand and distributed on the soil surface. The scheme of the treatments was as follows :

- in 1967 : - one foliar spray applied on 4. 4. 1967
  - composition of the mixture :

Form	quantity kg/ha	
	as salt %	as element
$MnSO_4 . 4H_2O$	20	1. 44
$CuSO_4 . 5H_2O$	20	1. 02
$MgSO_4 . 7H_2O$	19	0. 38
$ZnSO_4 . 7H_2O$	5	0. 23
$FeSO_4 . 7H_2O$	5	0. 20
$Na_2B_4O_7 . 10H_2O$	2	0. 05
$CoSO_4 . 6H_2O$	1	0. 05
NaCl	25	

- in 1968 : - two application of 10 kg respectively on 12. 4. 68 and 9. 7. 68
  - composition of the mixture :

	quantity kg/ha/application	
	as salt %	as element
$MnSO_4 . 4H_2O$	76.9	1.890
$ZnSO_4 . 7H_2O$	11.1	1.250
$CuSO_4 . 5H_2O$	5.1	1.130
$CoSO_4 . 6H_2O$	1.5	1.030
Borax $10H_2O$	5.4	1.015

2.4. Soil description and characteristics

The soil was classified as a sandy loam, and the different plots showed the following pH-values :

Plot	$pH-H_2O$	pH-KCl
O	5.5	4.1
NPK	5.5	4.1
NPKCa	6.2	5.0
NPKCaMg	6.2	5.2
-PKCa	6.6	5.4
N-KCa	6.6	5.4
NP-Ca	6.6	5.4

Mechanical analysis °

Sand %	36
Silt %	54
Clay %	10

Chemical analysis

C.E.C. 10.6. meq/100 g soil

° Data obtained from the "Centrum voor Grasland onderzoek" - Melle (Dir. Prof. M. SLAATS).

Trace element contents (extracted with O, 1 and O, 5 n HNO_3 - extraction ratio 1/5).

Trace element contents (extracted with O, 1 and O, 5 n HNO_3 - extraction ratio 1/5).

Treatment	Extraction	Fe	Mn	Zn	Cu	Pb
O	O. 1 n HNO_3	170	35.0	6. 2	2. 7	6. 5
	O. 5 n HNO_3	575	68. 8	17. 0	3. 0	20. 5
NPKCa	O. 1 n HNO_3	80	32. 5	5. 8	1. 0	3. 8
	O. 5 n HNO_3	760	85. 0	15. 5	2. 1	20. 5
-PKCa	O. 1 n HNO_3	100	36. 3	8. 0	1. 0	5. 0
	O. 5 n HNO_3	713	72. 5	15. 5	3.3	16.O
N-KCa	O. 1 n HNO_3	100	27. 5	3. 3	1. 0	3. 8
	O. 5 n HNO_3	345	40. 0	7. 0	3. 0	11. 3
NP-Ca	O. 1 n HNO_3	75	22. 5	5. 8	0. 8	4. 5
	O. 5 n HNO_3	563	47. 5	7. 0	3. 0	15. 3
NPK	O. 1 n HNO_3	100	30. 0	7. 3	1. 8	5. 3
	O. 5 n HNO_3	375	38. 8	7. 3	1. 9	9. 0
NPKCaMg	O. 1 n HNO_3	75	32. 5	5. 8	0. 8	5. 0
	O. 5 n HNO_3	713	62. 5	18. 8	3. 8	18. 5

- Organic matter (determined by the method of WALKLEY & BLACK)°

Treatment	% C			
	0 - 7 cm		15 - 20 cm	
	M. 39. 2	M. 39. 1.	M. 39. 2	M. 39. 1
1. Control	5. 3	3. 1	2. 1	1. 8
2. NPKCa	6. 4	3. 3	2. 3	2. 3
3. PKCa	5. 5	3. 3	2. 5	2. 2
4. N-KCa	5. 8	3. 1	2. 3	2. 3
5. NP-Ca	4. 5	2. 9	2. 0	2. 1
6. NPK	5. 4	3. 0	2. 2	2. 1
7. NPKCaMg	6. 3	3. 1	2. 4	2. 2

Climatic and weather conditions

Details concerning the distribution of the precipitation and maximum and minimum temperatures throughout the period of this study are given in tables below :

Table 23. 1 : distribution of the rainfull troughout 1967 and 1968 °

	1967		1968	
Month	mm.	days	mm	days
January	33	20	62	24
February	44	16	60	18
March	48	18	44	17
April	35	11	34	12
May	66	23	50	19
June	49	9	52	18
July	52	9	114	16
August	56	17	74	26
September	63	15	113	28
October	85	21	63	22
November	42	21	28	17
December	64	22	49	17
Total	637	202	743	234

° Data obtained from the Centrum voor Grasland onderzoek - Melle (Dir. Prof. M. SLAATS).

Table : 23.2 *

Year	Month	temperature mean daily max.	temperature mean daily min.	temperature highest values max.	temperature highest values min.
1967	January	5.8	1.3	13.6	- 8.3
	February	9.0	2.7	12.9	- 4.8
	March	11.0	3.9	15.9	+ 0.4
	April	12.6	2.7	17.7	- 3.1
	May	17.4	7.0	26.9	- 1.7
	June	19.7	9.4	25.0	+ 2.9
	July	23.9	11.8	32.6	+ 4.8
	August	22.2	11.5	29.6	+ 6.8
	September	19.0	10.6	24.8	+ 4.6
	October	15.4	8.9	20.6	+ 1.8
	November	8.1	2.0	12.4	- 6.0
	December	5.9	0.9	13.4	-13.0

Table : 23.3 *

Year	Month	temperature mean daily max.	temperature mean daily min.	temperature highest values max.	temperature highest values min.
1968	January	5.5	0.1	11.8	-15.6
	February	4.5	- 0.2	7.5	- 5.0
	March	10.7	+ 2.9	22.0	- 2.7
	April	15.0	3.1	27.0	- 4.7
	May	15.5	6.4	22.3	+ 0.1
	June	20.1	10.0	27.7	+ 3.1
	July	21.1	11.0	32.5	+ 4.6
	August	21.3	13.6	25.8	+ 8.6
	September	18.8	11.3	26.2	+ 9.0
	October	15.8	9.9	20.0	+ 4.6
	November	8.0	3.0	17.8	- 4.2
	December	3.0	- 2.0	11.9	-13.2

* Data obtained from the "Centrum voor Grasland onderzoek"- Melle (Dir. Prof. M. SLAATS).

2. 5. Botanical and chemical plant analysis

Separation between grass species and clover was carried out only during the year 1968 and the treatments selected for this purpose were no 3 (PKCa), 4 (N-KCa) and 7 (NPKCaMg). In 1967 the chemical analysis of the plants was carried out on mixed samples of the three replications. In 1968 each replication was separately analysed.

3. RESULTS :

3. 1. Dry matter production

Dry matter yields as influenced by major elements and trace elements application are given in table 24.

The influence of extra addition of trace element, on the dry matter yield is much less important than the influence of the respective different treatments with major elements comparing the rows a and b. It appears that the trace element application has increased the yield in some cases and decreased it in other cases. Summarising this observation, the influence of the trace element treatments was as follows, expressed in percentage values :

treatments	with trace elements	
	1967	1968
O	+ 7. 2 %	- 7. 8 %
NPKCa	- 1. 0	- 2. 3
PKCa	- 4. 5	+ 5. 8
NKCa	- 2. 2	- 7. 0
NPCa	+ 1. 1	+14. 6
NPK	+ 0. 7	- 2.0
NPKCaMg	+ 1. 7	- 1.0

These results indicate that any favourable influence of trace elements should be expected with regard to the chemical composition of the plants i. e. to their quality.

On the other hand the major element treatments had a very important influence on the dry matter production, and the lowest yields were systematically obtained on the control and on the plots where no potassium was applied.
The lack of phosphorous and of nitrogen was also acting severely. The highest yields were obtained on the plots with full fertilizations.

Table 24 : Dry matter yields (kg/ha) as influenced by trace elements and major element fertilizations.

	1967			1968		
Treatment	M. 39. 1. a	M. 39. 1. b	M. 39. 2	M. 39. 1. a	M. 39. 1. b	M. 39. 2
O	2. 781	2. 595	2. 123	2. 569	2. 786	3. 102
NPKCa	12. 714	12. 833	9. 747	11. 979	12. 263	10. 729
PKCa	10. 454	10. 945	9. 273	9. 215	8. 707	8. 491
N-KCa	8. 122	8. 301	4. 304	8. 364	8. 991	6. 131
NP-Ca	6. 393	6. 324	4. 812	7. 183	6. 266	4. 287
NPK	13. 154	13. 064	9. 181	12. 086	12. 323	10. 351
NPKCaMg	13. 125	12. 910	10. 118	12. 753	12. 867	10. 914

Dry matter yields (D. M. Y.) as influenced only by major element applications (M. 39. 2.) : The experiment indicated as M. 39. 2. recieved the same major element treatments as M. 39. 1, but no trace elements were applied. Table 24 shows that the D. M. Y. on these plots was generally lower than on the part M. 39. 1. Apart from this, the D. M. Y. in function of the different major element treatments follows the same order as already stated in experiment M. 39. 1. This means that the lowest yields were obtained on the control and the plots without potassium, followed by the treatment without phosphorus. The omission of nitrogen still gave higher yields than the treatments mentioned before, due to the fact that this resulted in a higher abundance of clover on the plots recieving -PKCa. Finally the full treatments with NPK, eventually completed with Ca and Mg, provided always the highest yields.

While the dry matter yield obtained in 1968 on part M. 39. 1. was lower than those obtained in 1967, the inverse was observed on the part M. 39. 2.

3\. 2. Effect of trace element application in different forms on trace element absorption

3\. 2. 1. Spray treatment : one dose

The effect of the trace element spray, applied in early april 1967, on the trace element content of pasture plants was rather small if we compare the treated and non treated parts a and b. However the first cutting after treatment showed a clearly pronounced increase in copper content, and the copper values went up till 146. 7 ppm (table 25 and fig. 18).

FIG. 18 EFFECT OF TRACE ELEMENTS FORTILIZATION (as spray) ON THE COPPER CONCENTRATION (P.P.m in dry matter) OF THE PLANT TISSUE

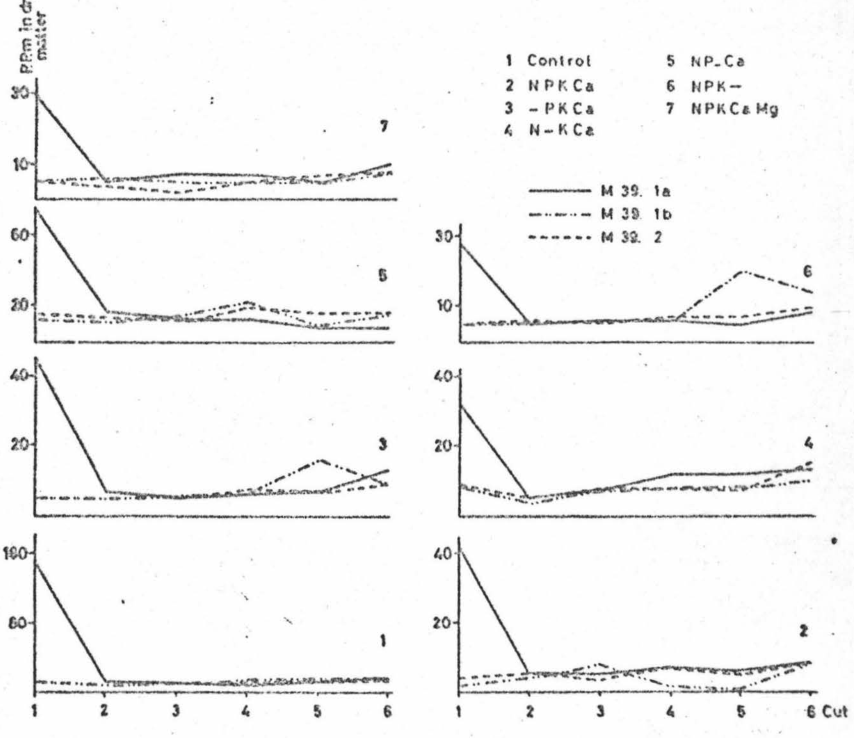


Table 25 : Effect of treated parts (a) and non treated parts (b) with trace elements (as spray) on the trace element concentrations (p.p.m. in D.M.) of the plant tissues during the first cutting.

Treat-ments		Fe	Mn	Al	Zn	Cu	B	Pb
0	a	414.3	1829	285.7	75.0	146.7	19.5	17.9
NPKCa	a	131.4	192.3	121.6	42.2	41.6	4.3	4.1
-PKCa	a	255.0	140.0	230.0	52.5	45.0	6.0	7.5
N-KCa	a	119.8	128.6	77.6	42.1	31.1	6.2	3.2
NP-Ca	a	439.3	346.8	242.8	63.1	73.2	16.2	12.8
NPK	a	122.0	153.5	108.2	42.3	27.5	3.5	3.7
NPKCaMg	a	133.6	152.1	113.0	42.1	29.0	3.2	5.1
0	b	298.1	1824.1	195.8	59.0	13.1	15.5	15.8
NPKCa	b	74.5	98.7	63.3	30.7	4.6	1.8	4.6
-PKCa	b	99.1	155.3	92.2	34.5	4.8	4.1	3.4
N-KCa	b	170.1	67.2	142.8	36.7	7.9	4.2	5.7
NP-Ca	b	364.5	255.5	182.2	57.4	12.1	18.2	13.0
NPK	b	85.5	152.0	74.1	34.2	5.1	3.0	3.6
NPKCaMg	b	86.8	93.7	68.6	33.3	5.0	2.8	4.0

As a matter of fact, burned leaves after spraying together with extremely high values of copper concentrations during the first cut were observed. This effect however was not observed after the first cut, and normal values in the different cuttings were obtained. The trace element concentrations of the plants were markedly influenced by the different major element treatments. The control as well as the treatment without potassium gave the highest trace element contents in the plants, followed by the treatment without nitrogen. This observation was also valid for the elements Fe and Cu, which are normally not subject to large fluctuations. Analogous observations were made with regard to the elements Al and Pb, though these elements were not present in the trace element mixture. Lower contents were generally found on the plots with full major elements fertilization (N-P-K, eventually combined with Ca and Mg). Analogous observations are valid for the part which did not recieve the trace element treatments, but there the same P effect did not appear as clearly, especially after the first cutting. In comparison to the parts without trace elements the average values of trace elements in the plants from the treated part are generally higher for the elements B, and Mn, but not for Fe, Cu and Zn. This phenomenon might be explained by the fact that the highest trace element concentration corresponds with the lowest yields, so that there seems to exist an enrichement of trace elements where the dry matter production remained low. Another possibility is linked with the deficiency of K, which might have been partially compensated by a higher absorption of cations in general and therefore also of the cationic trace elements. The validity of the latter possibility is supported by the figures obtained for Mg.
Indeed Mg reached a higher concentration level in the plants not treated with K(table 26), than in those treated with Mg and K together.

3.2.2. Application of trace elements as powder split into two doses :
As the high amounts of trace elements applied as a spray in 1967, resulted in toxic levels of copper, the application was made in 1968 as a powder mixed with sand, and the total amount was split into

Table 26 : Magnesium contents and uptake in function of major elements fertilisation.

Treatment	M.39.1.a			M.39.1.b			M.39.2		
	meq Mg per 100 g D.M.	% Mg	uptake in kg/ ha MgO	meq Mg per 100 g D.M.	% Mg	uptake in kg/ ha MgO	meq Mg per 100 g D.M.	% Mg	uptake in kg/ ha MgO
1. 0				16.4	0.20	8.56	22.3	20.27	9.55
2. NPKCa				10.1	0.12	25.67	10.8	0.13	21.44
3. -PKCa				12.2	0.15	27.36	12.8	0.16	25.28
4. N-KCa				9.8	0.12	16.60	9.2	0.11	10.08
5. NP-Ca				22.7	0.28	29.09	22.0	0.27	21.17
6. NPK	9.4	0.11	24.99	10.0	0.12	26.13	9.6	0.12	17.44
7. NPKCaMg	17.9	0.22	47.25	17.9	0.22	46.48	18.8	0.23	38.07

two doses. Since the pasture recieved no Fe in the trace element mixture during 1968, the Fe values obtained here will be mainly discussed in relation to the major element treatments and soil characteristics. The observations made in 1968 generally confirmed the findings of 1967. Plant analysis reveal the trace element treatments only in some particular cases, but once again the influence of the major element situations of the plots is showing a large influence on the tissue contents (table 28). In some cases the concentrations of trace elements, found when no fertilizers were applied and where K was omitted, run up to four or five times the contents in plants with full fertilization (tables 25, 27, 28, 29, 3O. 1 & 3O. 2).

Futhermore there were no significant differences between the trace element contents of the plants grown on plots with or without trace element fertilization.

Concerning Fe the mean values ranged between 116. 7 and 274. 4 p. p. m. and these variations correspond with the major element situation of the plots.

As to Mn, the mean values are between 81. 3 and 396. 8 p. p. m.

In this case the major element effect was accompanied by an effect of pH. Indeed, the lowest soil pH values were noticed on the control and on the N-P-K plots, and relative high Mn figures were also obtained on the latter plots.

The observed pH lowering was from 6. 6 to 5. 5 These observations are in agreement with GISIGER (195O), FIRGUS (1954) and others. On the other hand the lowest Mn values correspond with the treatments without nitrogen.

If we consider the Fe/Mn ratio in the plant tissues, a reversal of this factor was obtained in function of the major element fertilization, the full treatments NPKCa and NPKCaMg giving a ratio of nearly one. The other treatments comprising Ca and where no lowering the pH was noticed, provided as a rule plants with higher Fe than Mn contents.

Analogous high variations in Mn contents of different plants were also found by BEESON (1941) mentioning 4O to 936 ppm in lucerne, 79 to 51O ppm in red top grass as well as by HALE and HEINTZE

(1946) who found 30 to 500 ppm in green leaves. In our case the lowest and highest Mn values were obtained on the same plots during both years.

The mean Zn concentrations in the plants varied between 29. 97 and 59. 75, and as already stated, the highest values correspond with the control and the lack of potassium. As far as the other treatments concern, all the plants contained between 28. 62 and 33. 31 ppm of Zn, confirming the constancy of the Zn status of the plants. The same observation was made by MILLER et al. (1964), who also stated that the Zn content of herbage plants grown on a sandy loam soil, was not affected by either frequency or season of harvest.

The element copper behaves quite similarly as zinc and the same two plots also provided the highest copper containing plants (12. 15 till 16. 55 ppm). On the other plots the Cu concentrations ranged between 5. 99 and 9. 53 ppm. This means that no outstanding fertilizer effects could be noticed, which was also mentioned by HEMINGWAY (1962). It should be mentioned that the lowest values found, as well in 1967 as in 1968, are at the limit of requirement for a forage of satisfying quality. Our experimental scheme however did not permit to explain the low copper values by some trace element interactions, as reported by BOLL (1954). The boron figures reported in tables 27 & 28 also show quite large variations in function of the major element treatments, as well as in function of the successive cuttings and the season effect must be mentioned as an important factor in boron uptake.

Generally the first cutting gave the smallest boron contents, while the mean values ranged between 4. 70 and 25. 28 ppm.

This range corresponds with the values obtained by HANLEY (1962) who mentioned boron contents of 8 to 24 ppm for perennial ryegrass grown on 24 different soils with varying phosphate treatments.

Table 27 : Effect of treated (a) and non treated parts (b) with trace elements (as spray) on the trace element contents (mg/kg dry matter) and uptake (g/ha) - (values given as a mean over 6 cuttings).

Treatment		Fe		Mn		Zn		Cu		B		Pb	
		mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha
0	a + trace elements	201	560	1160	3226	49.2	137	23.9	67	16.1	45	8.5	24
NPKCa		94	1203	153	1942	33.8	430	10.6	134	3.6	45	2.5	32
-PKCa		216	2064	92	877	34.3	329	12.8	122	18.2	78	5.1	49
N-KCa		151	1222	81	660	30.1	245	10.2	83	6.0	49	3.0	24
NP-Ca		241	1541	219	1403	41.0	262	17.2	110	11.2	72	7.0	45
NPK		95	1245	151	1781	29.7	391	8.1	107	4.5	60	2.5	33
NPKCaMg		107	1399	132	1737	32.1	421	9.2	121	4.3	56	3.4	45
0	b - trace elements	210	546	1080	3841	48.7	126	11.7	30	23.7	62	9.5	25
NPKCa		111	1423	146	1783	29.7	381	7.0	90	3.3	53	3.0	38
-PKCa		132	1443	102	1113	30.5	334	6.0	66	10.6	116	3.7	40
N-KCa		117	970	94	781	22.7	189	6.7	56	2.9	35	4.3	36
NP-Ca		284	1796	281	1776	42.8	334	16.3	103	17.2	169	8.3	52
NPK		106	1386	180	2347	29.9	391	5.8	76	4.8	63	3.2	41
NPKCaMg		103	1329	126	1624	32.3	417	5.4	70	6.6	85	3.2	41

Table 28 : Effect of treated (a) and non treated parts (b) with trace elements (as a powder) on the trace element contents (mg/kg dry matter) and uptakes (g/ha) (values given as a mean over 6 cuttings).

Treat-ment		Fe		Mn		Zn		Cu		B		Pb	
		mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha
0	a +trace elements	275.6	708	1825	4688	58.3	149.7	14.5	37.2			11.8	30.5
NPKCa		146.5	1755	169.8	2034	31.4	375.5	8.3	99.4			5.2	62.2
-PKCa		190.5	1756	128.8	1187	29.1	268.7	10.5	82.3			6.4	59.8
N-KCa		182.7	1510	157.4	1301	31.0	256.2	10.5	87.0			4.8	39.5
NP-Ca		268.6	1929	183.0	1314	44.4	318.7	16.9	121.8			7.9	56.4
NPK		142.2	1719	263.7	3187	30.1	364.1	8.1	98.1			4.6	56.0
NPKCaMg		162.7	2075	137.9	1759	32.3	412.6	8.4	107.3			5.9	75.6
0	b -trace elements	234.8	654	1825	5084	58.5	163.2	13.2	37.0	22.0	61.4	12.9	36.2
NPKCa		152.1	1866	162.2	1189	24.8	305.2	8.5	104.3			4.4	54.2
-PKCa		171.6	1494	144.5	1258	29.3	255.7	9.2	80.1	8.2	71.7	6.2	54.1
N-KCa		168.6	1516	118.6	1066	28.8	259.3	9.3	83.9			4.0	36.5
NP-Ca		258.9	1685	208.3	1305	47.7	299.1	16.0	100.7	10.6	66.8	10.9	68.5
NPK		134.4	1656	247.8	2054	30.6	377.7	7.2	89.6			4.3	53.6
NPKCaMg		126.2	1624	133.6	1719	25.4	328.0	7.1	91.6			4.8	62.3

Table 29 : Effect of major element fertilisations (M.39.2) on the trace element contents (mg/kg of dry matter) and uptakes (mg/ha) (values given as a mean over 6 cuttings)

Treat-ment	Year	Fe		Mn		Zn		Cu		B		Pb	
		mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha
0	1967	208	442	780	1462	49.0	104	10.5	22	24.2	51	8.9	19
NPKCa		119	1163	167	1523	31.0	303	5.8	56			3.4	33
-PKCa		148	1437	87	843	28.0	273	6.3	62			4.8	44
N-KCa		131	693	98	522	32.2	171	8.0	42			3.1	17
NP-Ca		314	1509	243	1168	50.5	243	15.3	74	12.1	58	8.0	39
NPK		130	1189	153	1395	30.8	283	6.4	58			3.8	35
NPKCaMg		91	913	126	1258	29.5	296	4.9	50			3.0	30
0	1968	255.2	699	420.0	1303	55.1	171.0	12.5	38.9	[illegible]		9.80	30.4
NPKCa		154.1	1654	146.8	1575	31.2	334.4	6.4	69.1	[illegible]		4.85	52.0
-PKCa		191.6	1626	81.4	692	30.0	254.9	7.2	61.8			6.18	52.5
N-KCa		223.3	1369	103.9	637	35.1	215.2	11.2	68.9			5.74	35.2
NP-Ca		274.4	1176	171.4	735	40.7	174.3	14.4	61.8	9.9	42.7	9.24	39.6
NPK		117.2	1223	153.8	1592	29.7	308.1	5.9	61.5			3.87	40.1
NPKCaMg		145.3	1585	141.2	1542	33.1	261.3	6.5	70.4			5.20	56.7

Tables 30.1 & 30.2 show the significant differences between the (A) and lack of potassium (E) treatments in comparison with the others.

Table 30.1 : Differences between the plots treated (a) with trace elements (as powder) during 1968 : values based on the trace element contents (p.p.m. in D.M.).

O	(A)	The letters between brackets are used for identification in the biometric analysis.
NPKCa	(B)	
-PKCa	(C)	
N-KCa	(D)	
NP-Ca	(E)	
NPK	(F)	
NPKCaMg	(G)	

	Fe	Mn	Zn	Cu	B	Pb
treated part (a)	EB 126**	FG 150**	AC 32,1**	EF 8,8**	AB 19,8**	AF 7,9**
	EF 124**	FC 143**	AF 32,0**	EB 8,8**	AD 18,6**	AD 7,8**
	EG 113**	FB 121**	AD 30,8**	EC 8,4**	AF 17,9**	AB 7,7**
	ED 86,2**	ED 116**	AB 30,4**	EG 8,3**	AG 16,9**	AG 6,8**
	EC 86,9**	FE 86,9**	AE 17,2**	ED 6,5**	AC 9,3**	AC 6,2**
	AB 115**	EG 63,0**	EC 15,0**	EA 2,1**	AE 7,9**	AE 3,5**
	AF 112**	EC 55,6**	EF 15,0**	AF 6,7**	AB 11,8**	EF 4,3**
	AG 102**	ED 13;7**	ED 13,7**	AB 6,7**	ED 10,7**	ED 4,2**
	AD 74,7**		EB 13,2**	AC 6,2**	EF 9,8**	EB 4,2**
	AC 58,4**			AG 6,2**	EG 8,9**	EG 3,2**
	CB 56,6*			AD 4,4**	CB 10,4**	EC 2,7**
	CF 53,8*			DF 2,3**	DC 9,3**	CF 1,6**
				DB 2,3**	CF 8,5**	CD 1,5**
				DC 1,8**	CG 7,6**	DB 1,4**
				DG 1,8**		CF 1,1**
						BG 0,9*
non treated part (b)	EF 132**	EC 124**	AD 29,9**	EB 7,9**		AD 8,9**
	EG 120**	EF 113**	AB 29,6**	EF 7,7**		AF 8,8**
	EB 99,9**	EB 99,9**	AC 29,4**	EC 7,4**		AG 8,4**
	ED 85,0**	EA 83,1**	AF 29,2**	EG 7,4**		AG 7,7**
	EC 83,0**	ED 31,8	AG 28,4**	ED 5,7**		AC 7,0**
	AF 105**	DC 92,4**	AE 10,6**	AB 6,5**		AE 2,6**
	AG 93,6**	DF 82,2**	ED 19,3**	AF 6,3**		ED 6,3**
	AB 72,7**	DB 67,2**	EB 19,0**	AC 6,3**		EF 6,1**
	AD 57,6**	DA 51,3**	EC 19,0**	AG 6,0**		EB 5,8**
	AC 55,7*		EF 18,6**	AD 4,3**		EG 5,1**
	DF 49,3*		EG 18,0**	DB 2,2*		EG 4,3**
				DF 2,0*		CD 1,9**
				DC 1,9*		DF 1,8*
				DG 1,6*		CF 1,4*

* significant at 5 % ; ** significant at 1 %

Table 30.2 : Differences between the parcels treated with major elements (M.39.2.) during 1968 - values given as trace element contents (ppm in D.M.)

Fe	Mn	Zn	Cu	B	Pb
DF 279*	AC 458**	AF 24,6**	EF 7,2**	AF 14,0**	AF 5,3**
DG 234*	AD 434**	AB 23,5**	EG 6,8**	AB 13,6**	AB 4,5**
DB 231*	AG 396**	AC 22,7**	EB 6,4**	AG 12,1**	AG 3,8**
DC 219*	AF 396**	AG 21,0**	EC 5,8**	AD 10,6**	AD 3,7**
	AB 380**	AD 20,7**	ED 2,3*	AC 9,2**	AC 3,1**
	AE 370**	AE 15,5**	AF 6,4**	AE 8,6**	AE 1,1**
		EF 9,0**	AG 6,0**	EF 5,3**	EF 4,2**
		EB 8,0*	AB 5,7**	EB 3,9**	EB 3,4**
		EC 7,2*	AC 5,0**	EG 3,4**	EG 2,6**
				CF 4,7**	ED 2,5**
				CB 3,3**	EC 1,9**
				CG 2,8**	
				DF 3,4**	

3. 3. Further remarks :

3. 3. 1. Influence of botanical composition

In the preceeding paragraphs the results were given for the total herbage which is composed of different species as indicated under 2. 5. If one considers separately grass and clover, the analytical figures confirm that some trace element are not absorbed in the same proportions. Table no 31 gives the separate content of both these crops. From these figures it appears that the elements Fe, B and Pb are present in higher concentrations within the clover, while grass contains more Mn and no systematic difference is observed for Zn and Cu. Therefore the botanical composition of any herbage sward is a factor of importance in connection with its trace element situation. The same observations are valid as well as for the plots treated (a) and non treated (b) with trace elements.

Table 31 : The effect of botanical composition of the trace element contents (p. p. m. in D. M.) as a mean of three cuttings

Treatment	Species	Fe	Mn	Zn	Cu	B °°	Pb
PKCa a	grass	135. 8	113. 2	25. 4	8. 9	4. 7	4. 3
	clover	190. 0	68. 7	21. 9	6. 1	15. 1	10. 2
b	grass	122. 2	176. 7	26. 5	8. 6	4. 7	4. 8
	clover	157. 7	81.0	22. 8	5. 5	12. 6	6. 9
N-KCa a	grass	132. 5	79. 3	23. 4	7. 9	7. 0	3. 9
	clover	-	-	-	-	-	-
b	grass	130. 2	102. 3	27. 58	7. 9	4. 1	4. 1
	clover	-	-	-	-	-	-
NPKCaMg	grass	113. 6	88. 5	26. 1	6. 9	4. 6	4. 4
a	clover°	176. 1	82. 5	25. 7	8. 9	15. 1	8. 1
b	grass	115. 5	96. 3	25. 8	5. 9	4. 2	4. 5
	clover	147. 4	67. 0	23. 7	6. 9	16. 6	8. 6

a - recieved trace elements
b - recieved no trace elements

°° - for grasses, values of one cut were available
- for clover, values of two cuttings were available

3.3.2. Season effect

The results obtained during both years, show a season effect illustrating the evaluation of the trace element uptake from spring till autumn.

In a general way the first and the last cutting show the highest trace element contents and during the whole season some peaks also appeared when the dry matter production was lower. This may partly be explained as an effect of dilution by the organic matter production, but is not excluded that also other factors may have acted. So OZANNE (1955 b) mentioned that subterranean clover took up more zinc during long than during short day periods.

4. SUMMARY AND CONCLUSIONS

The influence of long term one-sided fertilization on the trace element absorption by the pasture plants was investigated. Changes in soil pH with regard to the different major element fertilization were notable. Indeed the lowest soil pH values were noticed on the control and on the NPK plots, and relative high Mn figures were also obtained on the latter plots.

High response of trace elements in the plants was recorded when these elements were added as spray ; this resulted particularly in toxic levels of copper.

The treatments giving the lowest dry matter productions systematically showed the highest trace element concentrations. Fluctuations in trace element contents due to seasonal variations were recorded.

From the botanical point of view, white clover contained higher values of Fe, B and Pb than grass, while the grass contained more Mn and no systematic differences were observed for Zn and Cu.

A. 2. LONG TERM FIELD EXPERIMENT (Since 1959)

1. INTRODUCTION :

Another analogous experiment called T. 59. 6. was organised in Merelbeke at "The Government Plant Breeding Station" in 1959 and this trace element study was undertaken for the harvests of 1967 and 1968.

2. EXPERIMENTAL DETAILS :

2. 1. Cropping history

The soil is classified as a moderate humid light sandy loam (Pdcz), with looze structure of B horizon.
In 1959 a grass-clover mixture was sown, composed of the species given in table 32.

Table 32 : Seed mixture sown on T. 59. 6. (kg/ha)

Species	T. 59. 6.
Lolium perenne L., perennial ryegrass pasture type C. V. 'Vigor'	5
Festuca pratensis Huds., meadow fescue C. V. 'Merbeem'	15
Pleum pratense L., Timothy, C. V. 'Erecta R. v. P.'	10
Poa pratensis L., Smooth stalked meadowgrass, C. V. 'Mervel'	3
Trifolium Repens L., White clover, C. V. 'Blanca R. v. P.'	3

Each year the plots were mown five times and during 1967 and 1968 two replicates were analysed for mineral elements.
During these two years the temperature and rainfall were noticed and given in preceeding chapter.

2.2. Fertilizer treatments

Eight treatments were applied on plots of 8 x 5 m with four replications under mowing conditions.
These treatments are as follows :

1. Control	no fertilizers
2. -PKCaMg	no N
3. N-KCaMg	no P
4. NP-CaMg	no K
5. NPK-Mg	no Ca
6. NPKCa	no Mg
7. NPK	no Ca-Mg
8. NPKCaMg	full fertilization

The fertilizer dressings applied yearly from 1959 were as follows :

Table 33 : Major element fertilizers applied to T. 59. 6 kg/ha/year

Treatment	N	P_2O_5	K_2O	Ca	Mg
Control	O	O	O	O	O
N-P-K-Ca-Mg	200 (1)	135 (2)	300 (3)	375 (4)	187. 5 (5)

Time of application

(1) N : nitrogen was applied at five applications of 40 kg/ha each

(2) P_2O_5 : 60 kg/ha before the first cut
40 kg/ha before the 3 cut
35 kg/ha before the 5 cut

(3) K_2O : 140 kg/ha before the first cut
100 kg/ha before the 3 cut
60 kg/ha before the 5 cut

(4) Ca : 750 kg/ha for two years, as $CaCO_3$

(5) Mg : 375 kg/ha for two years, as $MgSO_4$

3. RESULTS AND DISCUSSIONS

3.1. Soil

The chemical characteristics of the soil indicate a C.E.C. of 11.2 meq/100 g and a carbon content of 2.4 % as a mean value (Walkley & Black method).

The pH as influenced by the treatments reached in 1968 the following values on the different plots

Treatment	% C g/100 g of dry soil	pH H_2O	pH KCl
O	2.3	5.7	4.9
-PKCaMg	2.5	6.8	6.1
N-KCaMg	2.3	7.0	6.3
NP-CaMg	2.4	7.0	6.5
NPK-Mg	2.4	6.1	5.3
NPKCa	2.6	7.0	6.2
NPK	2.6	6.2	5.3
NPKCaMg	2.3	6.8	6.2

The major element situation of the different plots was also reflecting a measurable differenciation under the influence of the treatments and an extraction with Am-acetate (pH 4.8) gave the values shown in table 34.1.

Table 34. 1. : Extractable major elements (meq/100 g of soil) as extracted by Am-acetate (pH 4. 8) with relation to the major elements applied

Object	Ca	Mg	K	Na	P
	meq/100 g soil	meq/100 g soil	meq/100 g soil	meq/100 g soil	meq/100 g soil
Mowed					
1. O	4. 80	0. 43	0. 20	0. 11	1. 18
2. -PKCaMg	7. 52	0. 77	0. 28	0. 11	4. 22
3. N-KCaMg	7. 86	0. 73	0. 22	0. 12	0. 89
4. NP-CaMg	9. 40	0. 59	0. 12	0. 09	5. 34
5. NPK-Mg	5. 82	0. 64	0. 19	0. 15	2. 62
6. NPKCa	9. 13	0. 29	0. 16	0. 14	2. 42
7. NPK	6. 38	0. 27	0. 23	0. 14	2. 99
8. NPKCaMg	7. 90	0. 64	0. 20	0. 11	3. 26

In order to appreciate these figures, the following norms are to be taken into account [According to VAN DEN HENDE & COTTENIE (1960).]

Table 34. 2

Soil texture	Order of C. E. C. meq/100 g soil	classes	Meq extracted per 100 g soil		
			Ca	Mg	K
Light	5	very high	> 3	> 0. 5	> 0. 25
		high	3 - 2	0. 5-0. 2	0. 25-0. 15
		low	1-0. 5	0. 2-0. 1	0. 15-0. 08
		very low	0. 5	< 0. 05	< 0. 04
Medium	15	very high	> 10	> 1. 5	> 0. 4
		high	10-7. 5	1. 5-1. 0	0. 4-0. 3
		medium	7. 5-5	1. 0-0. 6	0. 3-0. 2
		low	5-2. 5	0. 6-0. 4	0. 2-0. 06
		very low	< 2. 5	< 0. 4	< 0. 06
Heavy	25	very high	> 20	> 2. 5	> 0. 8
		high	20-15	2. 5-2. 0	0. 8-0. 4
		medium	15-10	2. 0-1. 2	0. 4-0. 2
		low	10-5	1. 2-0. 6	0. 2-0. 1
		very low	< 5	< 0. 6	< 0. 1

The trace elements were determined in the same samples using O. 1 and O. 5 n HNO_3 as an extractant with soil-solution ratio of 1:5. The contents expressed in p. p. m. in the air dry soil are given in table 35.

Table 35 : Trace element extractability in relation to the different major element treatments applied.

ppm in air dry samples

		Fe	Al	Mn	Zn	Cu	Pb	Co	Ni
Control	0.1 n HNO_3	162.5	225	42.5	10.5	2.8	5.5	1	1.0
	0.5 n HNO_3	>1200	438	105	17.3	5.8	19.5	traces	4.0
- N	0.1 n HNO_3	150	238	45.0	10.8	4.8	5.0	1	1.0
	0.5 n HNO_3	>1200	500	120	21.4	6.3	37.0	traces	4.3
- P	0.1 n HNO_3	137.5	225	38.8	6.5	2.0	2.3	0.25	tr.
	0.5 n HNO_3	>1200	525	120	23.5	6.50	20.0	traces	5.3
- K	0.1 n HNO_3	200.0	255	45.0	19.5	2.5	5.8	1.50	1.5
	0.5 n HNO_3	>1200	538	134	29.3	4.8	22.8	traces	5.3
- Ca	0.1 n HNO_3	182.5	255	41.3	12.3	2.0	4.5	0.25	tr.
	0.5 n HNO_3	>1200	450	133	32.0	4.5	22.5	traces	1.0
- Mg	0.1 n HNO_3	137.5	250	52.5	7.3	3.0	4.5	1.50	1.5
	0.5 n HNO_3	>1200	490	114	15.8	6.5	13.8	traces	2.5
- Ca - Mg	0.1 n HNO_3	155.0	250	38.8	9.8	2.8	5.0	0.05	0.5
	0.5 n HNO_3	817.5	338	72.5	9.5	4.3	14.3	2.25	<1
full fertilisation	0.1 n HNO_3	125.0	230	38.8	5.5	2.8	5.0	0.05	1.5
	0.5 n HNO_3	>1200	600	113	17.8	8.3	26.8	traces	1.3

In order to judge these values it is useful to mention that the following mean figures are characteristic for the same soil group : (extraction with O. 5 n HNO_3) °

	Fe	Al	Mn	Pb	Zn	Cu
lowest values	88	225	7	1. 0	1. 5	0. 5
mean values	590	577	42	20. 5	20. 0	7. 5
highest values	1300	1240	115	55	28	18

Climatic and weather conditions

Details concerning the distribution of the precipitation and maximum and minimum temperatures throughout the period of this study are given in tables 36. 1, 36. 2 and 36. 3 .

Table 36. 1 : distribution of the rainfall throughout 1967 and 1968.

	1967		1968	
Month	mm.	days	mm.	days
January	34. 8	16	64. 4	25
February	49. 1	10	64. 5	18
March	48. 2	20	45. 5	17
April	38. 1	13	34. 9	13
May	96. 6	21	50. 5	20
June	39. 0	9	50. 2	17
July	44. 0	11	11. 3	18
August	59. 7	17	99. 1	23
September	72. 1	18	128. 1	24
October	89. 0	25	64. 5	19
November	43. 5	21	30. 3	12
December	66. 8	22	57. 2	15
Total	680. 9		800. 5	

° No sufficient figures are actually available for Co, Mo and Ni.

Table 36.2

Year	Month	Average t°			absolute extreme	
		month	max.	min.	max.	min.
1967	January	3.7	5.7	1.8	13	- 6
	February	6	8.7	3.4	12.8	- 3
	March	7.5	10.6	4.4	15.5	1
	April	7.8	12	3.6	16.8	- 0.5
	May	12.8	17.1	8.6	27	- 1
	June	14.1	17.3	10.9	25	5.5
	July	18.6	23.7	13.5	32.2	6.5
	August	17.6	22.4	12.9	24.5	9
	September	14.7	18.6	10.8	24.5	5
	October	12.4	15.4	9.4	20.5	3
	November	5.2	8.1	2.4	11.5	- 4.5
	December	2.9	5.5	0.4	13	-12.5

Table 36.3

Year	Month	Average t°			absolute extreme	
		month	max.	min.	max.	min.
1968	January	2.5	5	0.1	11.5	-15.5
	February	2.4	4.8	0.1	9	- 5
	March	6.5	9.7	3.3	22	- 2.5
	April	9.8	15	4.6	27	- 3
	May	11.1	14.9	7.4	22	2
	June	15.5	19.8	11.2	27	4
	July	16.7	21	12.4	32.5	7
	August	17.8	21.3	14.4	26	9.4
	September	15.1	18.8	11.4	26.5	9
	October	13.1	16.1	10.1	20	6.5
	November	5.8	8.4	3.2	20	- 3.5
	December	0.9	3.2	- 1.4	12	-13.2

3.2. Dry matter yields

The D.M. yields obtained in kg/ha are totalised in table 37.

Table 37 : Dry matter yields (D.M.Y.) kg/ha with relation to the major applications.

No	Treatments		1967	1968	Total
1	O - O - O - O - O	O	6.917	8.126	15.043
2	O - P - K - Ca - Mg	- N	8.973	8.597	17.570
3	N - O - K - Ca - Mg	- P	9.278	10.302	19.580
4	N - P - O - Ca - Mg	- K	8.147	9.570	17.717
5	N - P - K - O - Mg	- Ca	10.077	11.995	22.072
6	N - P - K - Ca - O	- Mg	9.368	11.087	20.725
7	N - P - K - O - O	- Ca - Mg	9.684	11.545	21.229
8	N - P - K - Ca - Mg	full	9.606	10.616	20.222

It is typical that the highest yields correspond with full fertilization, without calcium, that means with the plots showing a pH-H_2O of 6.1. In comparison with the control this treatments produced 46.7 % more D.M. during 1967 and 1968. Furthermore the plots without N and without K produced lower yields than the other treatments while the omission of phosphorus is also clearly appearing. These observations show a good parallelism with the results of the formerly mentioned soil analysis.

Finally a favourable effect of Mg, as already indicated, was marked when this element was applied in combination with calcium (full fertilizer treatment). Summarising these observations the D.M. showed the following very similar range over the two years of experimentation :

Year	Treatments							
1967	1	4	2	3	8	6	7	5
	O <	- K <	-N <	- P <	full fertilized <	- Mg <	- Ca - Mg <	- Ca
1968	1	2	4	3	8	6	7	5
	O <	- N <	-K <	- P <	full fertilized <	- Mg <	- Ca - Mg <	- Ca

3.2.1. Trace elements

3.2.1.1. Iron and manganese

In the case of iron, significant differences in herbage contents were only found in 1968 (table 39). The highest values correspond systematically with the treatment without potassium.
These results support the previous finding of the field trials M. 39. 1 and M. 39. 2. On the other hand there was a large difference between the Fe contents found in 1967 (157 to 225 p. p. m.) and 1968 (347 to 660 p. p. m.).
At the same time there was also a marked season effect, but this followed a different profile in both years. Indeed during 1967 a constant increase was observed from cutting to cutting, while in 1968 a net peak appeared with the cutting of August (fig. 19). The manganese contents were also higher in 1968 than 1967, but the difference was less pronounced than in the case of iron.
In the case of manganese however there was a highly significant influence from the major element treatments, the observations of both years confirming each other. The highest contents were found to correspond with the control (no fertilizer), the lower ones being correlated with the control of potassium, this as precisely with the plots given high Fe figures.
As the different treatments have resulted in quite pronounced pH variations, it also appears that the manganese contents closely correlated with this factor, the highest Mn values being linked to the lowest pH values and vice versa (fig. 23). Considering the Fe/Mn ratio, a systematic influence of the treatments with major elements was observed during both experimental years. Except in

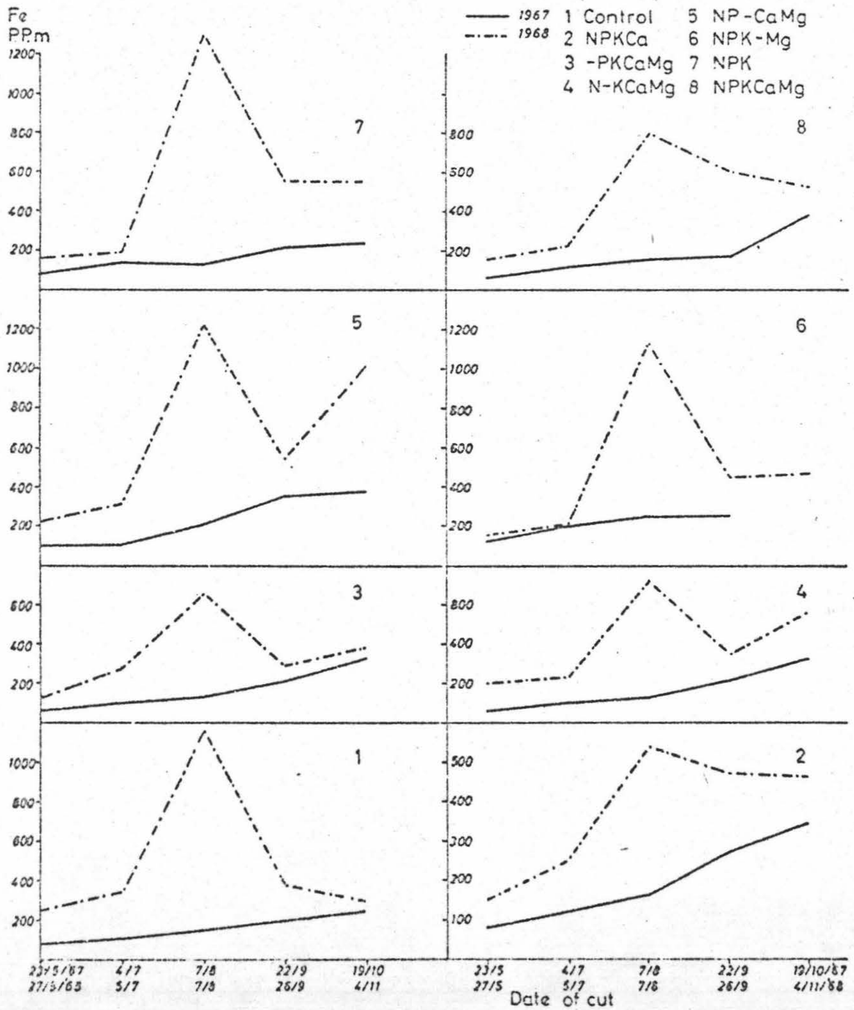


FIG. 19 THE INFLUENCE OF SEASONAL EFFECT ON THE IRON CONCENTRATION (P.P.m in dry matter) WITH RELATION TO DIFFERENT MAJOR ELEMENT TREATMENTS

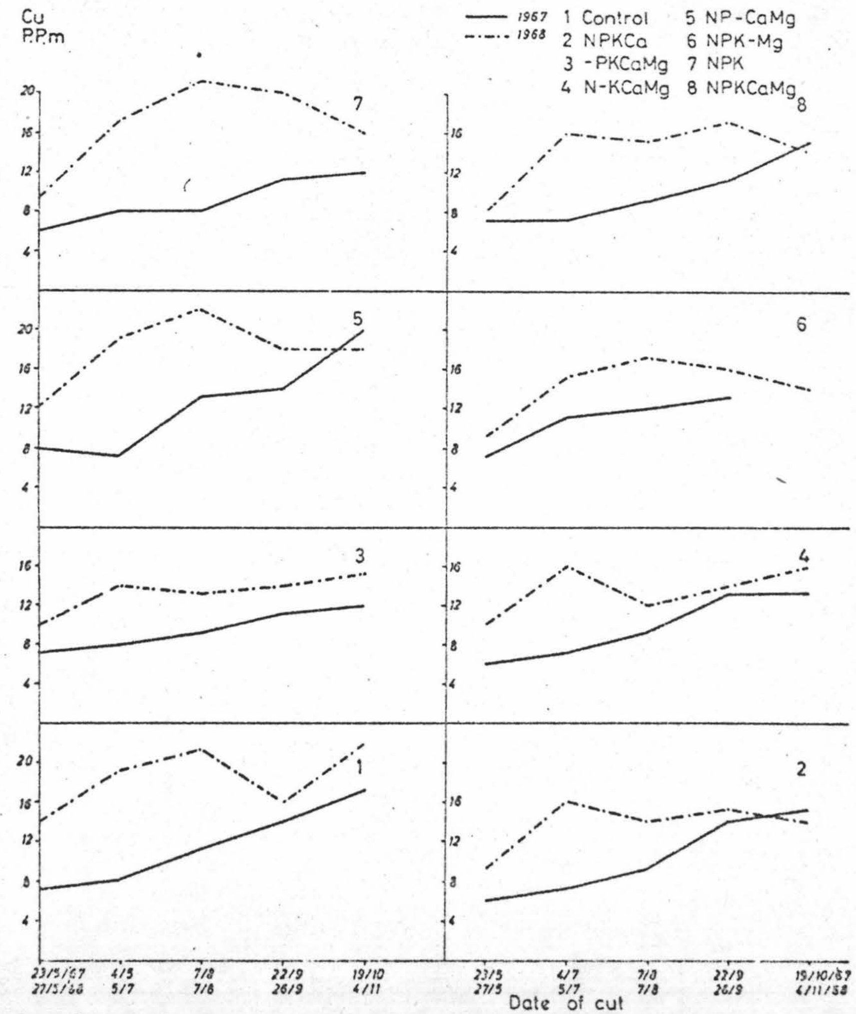


FIG. 21 THE INFLUENCE OF SEASONAL EFFECT ON THE COPPER CONCENTRATION (P.P.m in dry matter) WITH RELATION TO DIFFERENT MAJOR ELEMENT TREATMENTS

the control where the Fe/Mn ratio was equal to 1 or lower than 1, its values indicate normally 2 to 5 times more iron than manganese, the highest values being found where no potassium was applied. The soil itself contained 4 to 5 times as much extractable iron than manganese. The antagonism between these two elements was already reported by SOMERS and SHIVE (1942) and MORRIS and PIERRE (1947). This antagonism was apparently acting less in the control and most in the absence of potassium. The mechanism through which the Fe/Mn ratio was changing seems also to be linked with the pH variations of the soil, indeed this factor changed from 7 on the plots without potassium to 5. 7 on the control plots. Therefore it may be stated that a pH decrease favours relatively more the mobility of manganese than that of iron.

3. 2. 1. 2. Zinc

As in the case of iron, the zinc concentrations in the plant showed more variation in 1968 than in 1967. The mean values for this element reached the highest peak when no potassium was applied and low figures systematically correspond with the emission of manganese.

In spite of the fact that zinc did not give many statistical differences in function of the treatments (table 39), its concentrations varied in 1967 from 58. 4 to 119. 8 p. p. m. and in 1968 from 91. 5 to 187. 7 p. p. m.

3. 2. 1. 3. Copper and boron

Both the elements copper and boron were present at similar levels in the plants of this experiment. The seasonal variations of copper were however lower than of boron ; especially in 1967 there was a continuous increase of the copper content from spring to autumn. The most striking point is once again the fact that the control, as well as the plots without potassium treatment, gave the highest values for both elements (table 38). This observation shows that in spite of the fact that copper concentration is considered to be quite constant, some major element effect may however occur.

Moreover the plants grown in 1968 were higher in copper content than those harvested in 1967.
All the observed copper concentrations fall within the limits of safety for animal nutrition. The boron figures might on the other hand correspond with the critical values, if one takes into account the critical levels reported by various investigations and ranging between 5.8 and 23 p.p.m. (ROGERS 1947). In the present experiment the ranges of boron content were 6.27 to 18 p.p.m. in 1967 and 6.34 to 20.8 p.p.m. in 1968. A similar range (8-24 ppm) in perennial ryegrass was also reported by HANLEY (1962).
Finally table 38 also mentions the contents of lead in the plant of this experiment. They show the same trend as already reported for the other elements, particularly concerning the control and the treatment without potassium.

3.3. Total uptake

The total uptake of the different trace elements under study is also mentioned in table 39. With regard to these figures the differences in dry matter production often reversed the differences in trace element concentrations. Therefore the total uptake on the control plots is in the medium range, while the total uptake in generally the highest on the plots without calcium and/or magnesium treatments. In spite of medium dry matter yields on the plots without potassium treatment, the total uptake of trace elements remains high, due to their high contents in the corresponding plants.

3.4. Season effect

Various climatic factors have been found to affect the uptake of trace elements, and therefore their concentration in the plant. These factors however were clearly pronounced in the present study and showed that the concentration of the elements given in table 38 were appreciably higher in 1968 than in 1967. Another important point is the fact that during the period of August 1968, the accumulation of Fe, Mn and Zn reached up to a maximum (fig. 19, 20 & 21). Also copper showed an analogous tendency (fig. 22).

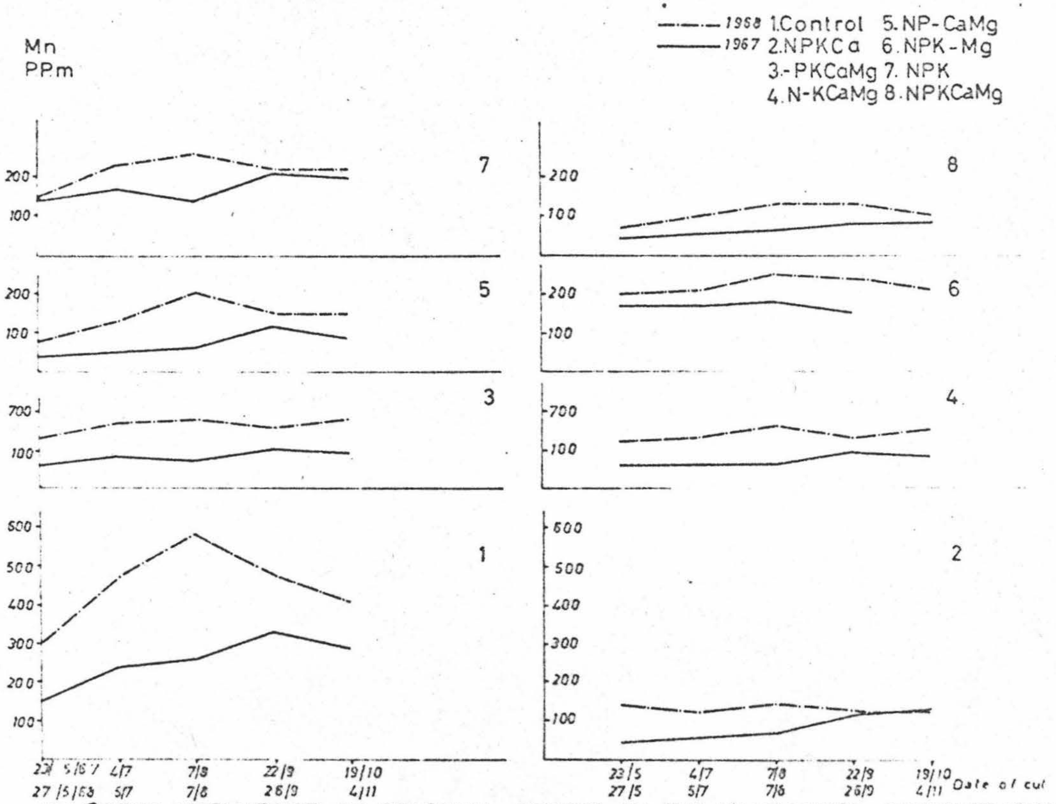


FIG.20 THE INFLUENCE OF SEASONAL EFFECT ON THE MANGANESE CONCENTRATION (PPm in dry matter) WITH RELATION TO DIFFERENT MAJOR ELEMENT TREATMENTS

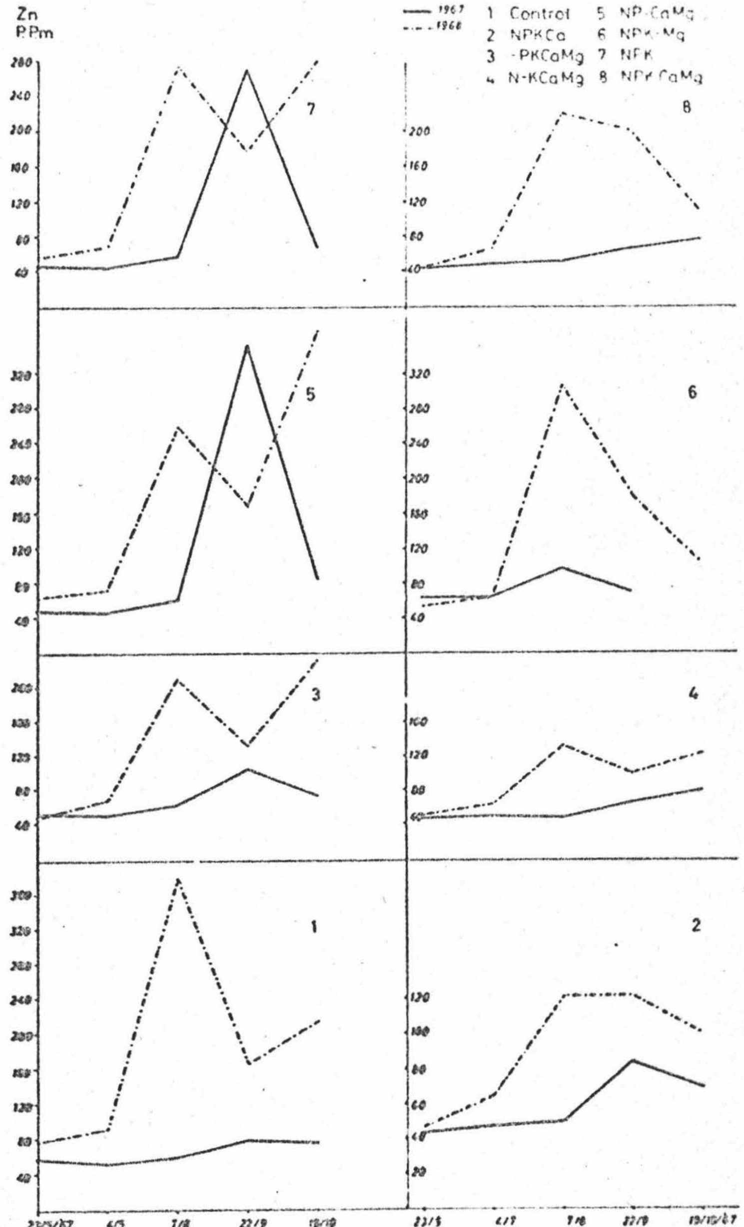
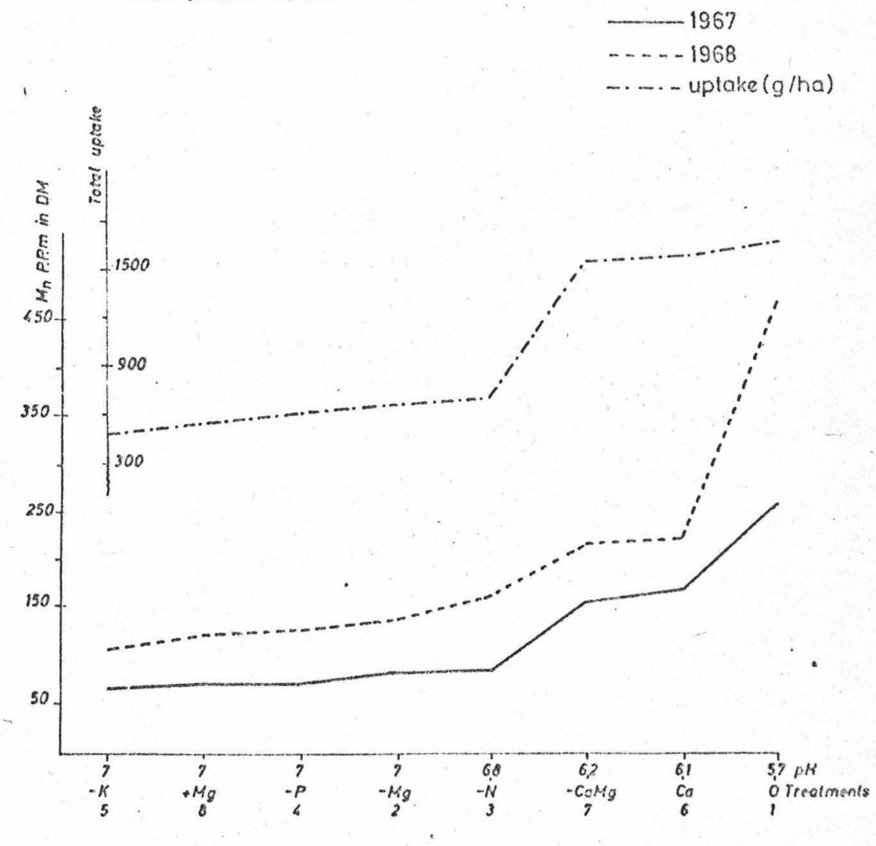


FIG. 22 THE INFLUENCE OF SEASONAL EFFECT ON THE ZINC CONCENTRATION (P.Pm in dry matter) WITH RELATION TO DIFFERENT MAJOR ELEMENT TREATMENTS

FIG. 25 RELATION OF MANGANESE TO CHANGES IN pH OF SOIL [redacted]



In order to explain this phenomenon, it is necessary to draw the attention to the tables 36. 1 ; 36. 2 and 36. 3 given in pages 96 and 97, where the amount of rainfall and the temperature during both years is given. These tables emphasise an increase of rainfall and temperature during the same period.
Nevertheless, it appears that fluctuations in mineral contents due to soil moisture (rainfall) levels and temperature changes bring about many differences in plant composition besides those related to the availability of the nutrients in soils. Support to this observation was reported in different soil conditions by MC COOL (1934), LINDHARD (1948), DORPH-PETERSEN (1950), MAROCKE (1957), NOZDRUNOVA et al. (1958), HODGSON (1963) and others.

4. CONCLUSIONS

The field experiment T. 59. 6 indicates that after some ten years of repeated different treatments with major elements, the trace element contents and uptake by pasture plants were significantly differenciated for different elements. The most sensitive elements are manganese, responding to slight pH differences, as well as copper, boron and lead. Iron and zinc were less influenced by the treatments, but showed a more substantial seasonal effect.
The Fe/Mn ratio appeared to be a largely influenced factor. Finally the total uptake of trace elements showed a quite different pattern than the one obtained in considering only the concentrations of the same elements in the plant tissues.
It should be also mentioned that the contents of Fe, Al, Mn, Zn, Cu and B in the plants, were notably higher in 1968 than in 1967, and this in spite of higher dry matter yields in 1968. This means that the relationship between the nutrient level of the soil and the mineral content of the herbage, as observed by GESSEL (1970), is also related to factors such as seasonal variations, rates and combinations of fertilizers etc.

Table 38 : Effect of major element fertilisations (T.59.6) on the trace element contents (mg/kg of dry matter) and uptake (g/ha) : values given as a mean over 5 cuttings.

Object	Fe		Mn		Zn		Cu		B		Pb		Fe/Mn ratio
	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	
0	157.0	927	254.0	1659	64.90	421	11.00	66.33	18.00	114.0	5.17	33.55	0.62
-PKCaMg	176.0	1497	77.70	689	68.79	585	9.17	76.34	12.52	115.1	6.00	55.10	2.23
N-KCaMg	165.4	1233	67.53	595	58.36	498	9.52	80.21	9.26	81.46	5.29	43.31	2.43
NP-CaMg	225.2	1436	69.29	494	119.8	856	12.59	86.23	10.00	73.32	7.23	51.43	3.26
NPK-Mg	207.0	1875	116.2	1570	73.27	671	10.88	99.18	13.00	125.0	6.55	62.59	1.25
NPKCa	182.7	1572	79.00	664	59.44	543	9.97	86.83	8.74	73.51	5.53	54.36	2.44
NPK	162.9	1401	152.6	1542	97.36	893	8.99	80.37	9.17	90.51	4.68	44.43	1.07
NPKCaMg	182.4	1362	63.43	549	57.46	511	9.65	82.60	6.27	54.00	5.71	49.27	2.88
0	483.6	3682	467.2	3753	187.6	1306	18.26	136.9	20.77	160.0	8.14	57.51	1.04
-PKCaMg	347.5	2374	161.8	1306	138.9	902	13.10	104.0	10.70	77.93	6.10	44.48	3.15
N-KCaMg	495.4	3997	136.2	1332	93.79	840	13.46	129.0	9.75	88.15	6.26	56.71	3.64
NP-CaMg	660.1	5428	123.4	1170	187.7	1049	17.71	160.0	14.95	146.2	7.86	72.36	5.37
NPK-Mg	485.9	5052	220.2	2605	142.2	1559	14.29	160.0	10.99	110.5	6.48	69.39	2.21
NPKCa	375.6	3532	128.1	1435	91.45	904	13.63	140.3	6.34	62.76	5.72	56.10	2.94
NPK	551.0	5438	215.4	2343	173.3	1578	13.34	143.1	10.40	109.5	6.37	64.43	2.56
NPKCaMg	467.7	4369	107.9	1080	128.3	1237	13.72	135.5	9.99	96.82	6.49	63.14	4.33

Table 39 : Statistical evaluation of trace element contents of the plant tissues (T.59.6), as influenced by major element fertilisation during 1967 and 1968 : values given as a mean over five cuttings.

Year	Fe	Mn	Zn	Cu	B	Pb
1967		AH 190** AD 186** AE 184** AC 175** AB 174** AG 101* GH 98,1* GD 85,1* GE 83,3*		EG 3,60** EC 3,42** ED 3,07* EH 2,94* EB 2,62*	AH 12,6** AG 9,54* AB 9,26* AD 8,74* AE 7,97*	EG 2,55* EA 2,07* ED 1,94*
1968	EC 313* EB 285*	AH 359** AB 339** AD 331** AE 327** AC 305** AG 252** AF 247** FH 112** GH 107*	EB 96,3* ED 93,9* AB 96,2* AD 93,9*	AC 5,17** AG 4,91** AD 4,79** AB 4,63** AH 4,54** AF 3,96* EC 4,59** ED 4,22** EB 4,06** ED 4,22** EH 3,96* EF 3,39*	AB 14,4** AD 11,0** AH 10,8** AG 10,4** AC 10,1** AF 9,78** AE 5,82** FD 8,62** ED 5,22** EH 4,94** EG 4,56* EC 4,27* EF 3,95* FB 4,67* CB 4,35** GB 4,06* HB 3,68** DB 3,40*	AB 2,40** AC 2,06* AD 1,86* AG 1,76* AF 1,76* AH 1,64* EB 2,13* FC 1,78* FD 1,59* EG 1,49* EH 1,37*

* significant at 5 % ; ** significant at 1 %.

A O
B -Mg
C -N
D -P
E -K
F -Ca
G -Ca and -Mg
H full

the letters between the brackets are used for identification in the biometric analysis.

B. SHORT TERM FIELD EXPERIMENT

1. INTRODUCTION :

In order to observe the trace element uptake by pasture crops as influenced by short term differences in major element fertilization, two fields were selected on which different N-P-K fertilizers were applied since 1967. In 1968 a treatment with and without magnesium was added. Details of the layout are given below.

2. EXPERIMENTAL DETAILS :

2. 1. Site and cropping history :

Both experimental fields are located in Lemberge and following swards were used :

Table 4O : Seed mixture sown on T. 67. 1O and T. 67. 12

Species	mowed and grazed T. 67. 10	mowed T. 67. 12
Lolium perenne L., perennial ryegrass pasture type C. V. 'Vigor'	4	10
Lolium perenne L., perennial ryegrass hay pasture type R. v. P.	6	-
Festuca pratensis Huds., meadow fescue C. V. 'Merbeem'	12	12
Pleum pratense L., Timothy, C. V. 'Erecta, R. v. P.'	10	10
Poa pratensis L., smooth stalked meadow-grass, C. V. 'Mervel'	2	-
Poa trivialis L., rough stalked meadowgrass	2	3
Trifolium repens L. white clover, C. V. C. V. 'Blanca R. v. P.'	3	-
Trifolium pratense L. red clover, C. V. 'Violetta R. v. P.'	3	3
Trifolium hydridum L., swedish clover	-	4

2.2. The P and K fertilizer treatments were applied with four replications and repeated with two levels of nitrogen, as shown in the following scheme.

In the experiment T. 67. 10 the plots were mowed and the aftermath grazed. The field called T. 67. 12 was exclusively mowed.
The quantities and rates of this fertilization are as follows :

1967 : Fertilizers applied to T. 67. 10 and T. 67. 12 kg/ha/year

No	mowed + grazed T. 67. 10		mowed T. 67. 12	
	P_2O_5	K_2O	P_2O_5	K_2O
1	O	O	O	O
2	120	120	120	120
3	90	180 (i)	135 (ii)	300 (iii)

(i) : given in 2 applications : before the 1st cut
after the 1st cut

(ii) : given in 3 applications : before the 1st cut 6O kg
after the 1st cut 4O kg
after the 2nd cut 35 kg

(iii) : given in 3 applications : before the 1st cut 14O kg
after the 1st cut 1OO kg
after the 2nd cut 6O kg

Phosphate fertilizer was applied as fertifos (38 %) and Potassium as KCl 4O %.

1968 : Fertilizers applied to T. 67. 10 and T. 67. 12 trials, kg/ha/year

No	mowed + grazed T. 67. 10		mowed T. 67. 12	
	P_2O_5	K_2O	P_2O_5	K_2O
1	O	O	O	O
2	120	200	120	200
3	135	400	135	400 (iiii)

(iiii) : given in 3 applications : before the 1st cut 2OO kg/ha
after the 1st cut 12O kg/ha
after the 2nd cut 8O kg/ha

Remark : - In 1968 all the plots were divided into two parts, one part recieving 25O kg/ha of Mg in the form of Kiserite.

Nitrogen applied as two levels "low and high" combined with P and K as follows :

Amount of N fertilizer applied in kg N/ha/year

		Low N kg/ha	High N/kg/ha
T. 67. 10	mowed + grazed	110 (50+3x20)	220 (100+3x40)
T. 67. 12	mowed	150 (3x50)	300 (3x100)

2.3. Successive cuttings : In 1967 both fields were sampled single time, because this was the first year after sowing. In 1968 four cuttings were obtained from the part T. 67. 10 and three from T. 67. 12.

2.4. Soil characteristics :
The sandy loam soil where the experimental fields were situated had a carbon content of 0.8 to 1.0 % C. E. C. - of 5.3 meq/1OO g and the pH and the nutrient indexes of the different plots at the end of 1968 are given in tables 41. 1, 41. 2, 41. 3 & 41. 4 (same methods as described before).

Table 41. 1 : Extractable major elements (meq/100 g of soil) as extracted by Am. acetate (pH 4. 8) with relation to the major elements applied (T. 67. 10).

Treatment	P	K	Ca	Mg	Na
	mg/100 g soil	meq/100 g soil	meq/100 g soil	meq/100 g soil	meq/100 g soil
High N					
0P - 0K	1. 69	0. 09	3. 73	0. 37	0. 08
120P-120K	2. 66	0. 12	3. 98	0. 35	0. 08
90P-180K	2. 35	0. 15	3. 95	0. 39	0. 09
Low N					
0P - 0K	1. 54	0. 12	3. 78	0.42	0. 08
120P-120K	2. 64	0. 16	3. 84	0. 40	0.0 8
90P-180K	2. 14	0. 16	3. 95	0. 39	0.0 9

Table 41.2 : Extractable major elements (meq/100 g of soil) as extracted by Am. acetate (pH 4.8) with relation to the major elements applied (T.67.12).

Treatment	P	K	Ca	Mg
	mg/100g soil	meq/100g soil	meq/100g soil	meq/100g soil
High N				
0P-0K	1.38	0.07	3.53	0.25
120P-120K	2.57	0.11	3.86	0.26
135P-300K	2.29	0.12	3.64	0.36
Low N				
0P-0K	1.49	0.08	3.42	0.28
120P-120K	2.40	0.12	3.47	0.30
135P-300K	2.01	0.17	3.29	0.30

Other chemical characteristics of the soil are given as follows :

Trial no	Treatment		% C g/100 g of dry soil	pH H_2O	pH KCl
T.67.10	HN	0P-0K	1.1	6.8	5.4
	HN	120P-120K	0.9	6.6	5.6
	HN	90P-180K	1.0	6.6	5.5
	LN	0P-0K	0.9	6.7	5.3
	LN	120P-120K	1.1	6.4	5.7
	LN	90P-180K	1.0	6.3	5.4
T.67.12	HN	0P-0K	0.9	6.5	5.6
	HN	120P-120K	1.0	6.7	6.2
	HN	90P-180K	0.8	6.8	6.3
	LN	0P-0K	0.9	6.4	5.5
	LN	120P-120K	0.8	6.4	5.4
	LN	90P-180K	0.9	6.5	5.5

HN - high nitrogen ; LN - low nitrogen

Table 41.3 : Extractable trace elements in T.67.10 trial, with relation to the different major elements applied (ppm in air dry sample)

	Treatment		Fe	Mn	Al	Zn	Cu	Pb	Co	Ni
High N	0P-0K	0.1 n HNO_3	187.5	46.25	207.5	5.00	2.00	6.50	0.05	tr.
		0.5 n HNO_3	517.5	78.75	400.0	8.50	4.50	13.00	-	<1
	120P-120K	0.1 n HNO_3	187.5	40.00	200.0	8.50	3.00	12.50	2.00	2.75
		0.5 n HNO_3	650.0	57.50	280.0	10.50	4.25	16.50	-	1.25
	90P-180K	0.1 n HNO_3	130.0	12.50	187.5	20.50	2.00	4.50	tr.	tr.
		0.5 n HNO_3	462.5	46.25	237.5	20.00	3.50	15.75	-	<1
Low N	0P-0K	0.1 n HNO_3	130.0	26.25	185.0	5.50	2.00	5.50	0.05	0.25
		0.5 n HNO_3	517.5	78.75	450.0	7.75	4.75	13.00	-	<1
	120P-120K	0.1 n HNO_3	137.5	27.50	187.5	5.50	1.75	4.50	0.25	1
		0.5 n HNO_3	612.5	66.25	375.0	8.00	4.75	14.50	-	<1
	90P-180K	0.1 n HNO_3	125.0	30.00	200.0	6.75	2.00	5.25	0.25	tr.
		0.5 n HNO_3	525.0	48.75	262.5	6.40	3.50	12.25	-	<1

Table 41.4 : Extractable trace elements of T.67.12 trial with relation to the different major elements applied : p.p.m. in air dry sample

			Fe	Mn	Al	Zn	Cu	Pb	Co	Ni
High N	OP-OK	0.1 n HNO_3	100	38.75	250	5.75	2.50	4.25	1.25	1.00
		0.5 n HNO_3	662	77.50	335	2.25	3.75	16.25	-	<1
	120P	0.1 n HNO_3	100	37.50	255	6.50	3.00	4.50	1.00	0.75
		0.5 n HNO_3	530	71.25	450	11.50	5.75	14.00	-	<1
	135P-300K	0.1 n HNO_3	175	35.00	287	5.25	3.75	3.75	0.05	1.00
		0.5 n HNO_3	485	50.00	425	6.50	4.50	12.00	-	<1
Low N	OP-OK	0.1 n HNO_3	80	25.25	242	5.75	2.50	15.25	0.08	26.25
		0.5 n HNO_3	575	50.00	300	6.50	3.50	29.00	<1	50.00
	120P-120K	0.1 n HNO_3	112	27.50	220	4.50	2.00	4.50	0.08	27.50
		0.5 n HNO_3	612	66.25	375	8.00	4.75	14.50	-	66.25
	135P-300K	0.1 n HNO_3	130	27.50	225	5.00	2.00	5.00	tr.	27.50
		0.5 n HNO_3	625	75.00	487	9.50	6.00	55.00	-	75.00

As the field were located in the same area as experiment no. T. 59. 6, the climatic conditions given under A 2 are also valid in this case.

3. RESULTS AND DISCUSSION :

3. 1. Dry matter yield

The dry matter yield (total) obtained in 1968 are given in table 42.

Table 42 : D. M. Y. (kg/ha) as influenced by different levels of major elements fertilization (1968).

	Rates of P and K	T. 67. 10		Rates of P and K	T. 67. 12	
		- Mg	+ Mg		- Mg	+ Mg
Rates of N		yields kg/ha	yields kg/ha		yields kg/ha	yields kg/ha
High	0P-0K	10. 546	10. 876	0P-0K	9. 708	9. 574
	120P-120K	12. 314	12. 952	120P-120K	13. 822	13. 837
	90P-180K	12. 585	12. 769	135P-300K	13. 989	14. 452
Low	0P-0K	9. 069	8. 315	0P-0K	8. 575	8. 683
	120P-120K	11. 120	10. 867	120P-120K	12. 731	13. 039
	90P-180K	11. 315	10. 430	135P-400K	14. 664	13. 819

The results in table 42 show the positive effect of nitrogen. Phosphorus and potassium have also significantly increased the yields, but it is difficult to distinguish clearly between the different rates of these elements in experiment T. 67. 10. The large difference in potassium fertilization on the field T. 67. 12 however resulted in a significant yield increase.

3.2. Trace elements :

3.2.1. Sample preparation : As it seemed quite difficult to collect pasture samples without any contamination of soil, it was decided to compare the analytical results obtained by direct analysis of dried and finally graded samples with those after initially washing the samples with pure doubled distilled water. This experiment was carried out using the 1967 samples of field T.67.10. The results obtained for several trace elements are given in table 43.

Table 43 : The effect of washed and not washed pasture samples on the trace element concentrations (p.p.m. in dry matter) of the total shoots.

Element	W/NW
Al	°°
Fe	°°
Mn	°°
Cu	°°
Pb	°°

°° significant at 1 %

Statistical interpretation of these results confirm a highly significant difference between both preparation technics and lead to the decision of washing all samples before analysis.
This method was indeed applied from 1967 for all trace elements analysis.
The same problem was also mentioned by THOMPSON (1957).

3.2.2. Trace element absorption

Iron and Manganese : The iron concentration in the plants shows some significant differences in function of PK and Mg treatments compared to the control, this as well at the low nitrogen and high nitrogen level (see tables 46.1, 46.2, 46.3 and 46.4.
The same was the case with zinc, while all the other trace elements only gave significant differences with regard to the mean concen-

trations, not taken into account the nitrogen level.
Referring to experiment T. 59. 6, the manganese contents obtained here were much more constant. This corresponds with the fact that the different treatments did not give rise to large pH fluctuations. Indeed the pH values of all the plots at the end of the experiment ranged between 6. 4 and 6. 8.
This confirms our earlier statements with regard to the manganese uptake by plants.

Table 44 : Range of trace element contents in 1968 (ppm in dry matter)

	T. 67. 10		T. 67. 12	
	- Mg	+ Mg	- Mg	+ Mg
Fe	177. 9 - 216. 4	311. 6 - 314. 2	221. 2 - 472. 8	263. 0 - 769. 0
Mn	132. 8 - 172. 9	156. 4 - 195. 7	171. 7 - 264. 7	168. 7 - 257. 0
Zn	92. 51 - 145. 6	136. 7 - 203. 6	202. 0 - 450. 6	258. 7 - 476. 9
Cu	11. 70 - 15. 15	12. 73 - 17. 50	12. 47 - 18. 43	12. 52 - 19. 36
B	10. 42 - 12. 06	12. 21 - 18. 49	6. 18 - 11. 77	5. 50 - 9. 33
Pb	4. 64 - 5. 91	5. 55 - 7. 70	9. 13 - 13. 20	9. 56 - 14. 80

The magnesium treatments appear to have increased the contents of trace elements in the plants quite generally.
The total uptake of trace elements is also given in tables 45. 1 & 45. 2 and follows the same trend as the total production, at least when the effect of nitrogen as well as the comparison between the treatments and the controls is considered.
The differences in total uptake between treatments which did not result in substantial differences of yields are however mainly related to the formerly discussed differences in trace element concentration of the plant tissues.

Table 45.1 : Influence of major element fertilisation (T.67.10) on the trace element contents (mg/kg of dry matter) and uptake (g/ha) : values given as a mean over 4 cuttings.

	Object	rates of nitrogen	Fe		Mn		Zn		Cu		B		Pb		Fe/Mn ratio
			mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	
- Mg	0P-0K	high	216.4	1842	172.9	1889	145.6	1330	15.15	126.9	11.24	105.0	5.91	58.42	1.25
	0P-0K	low	199.8	1578	154.6	1426	110.8	675.1	15.69	103.3	10.42	81.0	5.18	47.41	1.29
	120P-120K	high	183.6	1944	145.4	1941	97.22	1025	12.62	138.3	10.50	119.8	4.97	59.13	1.23
	120P-120K	low	177.9	1826	142.4	1773	107.6	107.6	12.19	120.9	12.06	126.2	4.76	53.72	1.25
	90P-180K	high	192.7	2350	135.6	1781	105.8	1490	11.70	132.8	10.79	111.2	4.64	58.12	1.42
	90P-180K	low	191.6	1885	132.8	1665	92.51	1107	12.10	121.2	10.88	112.2	4.68	52.92	1.44
+ Mg	0P-0K	high	389.0	2717	193.8	1984	203.6	1426	17.50	140.0	18.49	182.5	7.70	74.43	2.06
	0P-0K	low	414.2	2257	195.7	1502	173.5	1050	15.83	107.9	15.33	100.3	6.84	51.17	2.25
	120P-120K	high	351.5	3146	164.0	2158	136.7	1372	13.93	142.5	12.21	133.2	6.26	67.76	2.15
	120P-120K	low	333.7	2097	190.2	2092	156.7	1615	13.00	128.0	15.95	146.5	5.66	59.17	1.76
	90P-180K	high	326.0	3034	156.4	2009	149.2	1581	14.10	176.1	13.56	142.2	5.80	64.00	2.09
	90P-180K	low	311.6	2471	177.0	1896	155.9	1319	12.73	117.0	13.83	130.1	5.55	54.42	1.76

Table 45.2 : Influence of major elements fertilisation (T.67.12) on the trace element contents (mg/kg of dry matter) and uptake (g/ha) : values given as a mean over 3 cuttings.

	Object	rates of nitrogent	Fe		Mn		Zn		Cu		B		Pb		Fe/Mn ratio
			mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	mg/kg D.M.	g/ha	
− Mg	0P-0K	high	472.8	3808	264.7	2419	450.6	4721	18.43	168.2	11.77	117.9	12.83	109.2	1.78
	0P-0K	low	328.2	2283	225.5	1886	316.3	2075	17.78	127.3	10.56	84.89	13.20	89.9	1.45
	120P-120K	high	374.8	4334	263.3	3365	429.0	5406	15.99	200.7	8.83	99.77	12.38	131.9	1.43
	120P-120K	low	221.2	2393	203.9	2707	202.0	2490	13.85	160.0	9.13	111.9	10.00	102.4	1.10
	135P-400K	high	312.0	3802	215.2	2936	384.9	5107	13.70	182.3	6.18	67.98	10.44	120.2	1.45
	135P-400K	low	222.0	2739	171.7	2436	255.2	2669	12.47	162.2	7.23	95.80	9.13	104.7	1.29
+ Mg	0P-0K	high	769.0	5611	257.0	2376	461.7	4703	19.36	168.9	9.25	87.50	14.80	119.3	2.99
	0P-0K	low	404.0	2584	214.7	1769	467.9	3747	17.00	122.7	8.84	71.30	13.27	89.8	3.23
	120P-120K	high	320.3	3720	206.0	2753	427.7	6168	14.77	192.2	6.26	72.08	11.42	125.8	1.55
	120P-120K	low	386.0	3200	197.7	2582	287.6	3467	14.77	168.7	9.32	118.7	11.24	119.5	1.44
	135P-400K	high	294.0	3740	194.0	2829	323.8	4774	14.37	194.6	5.50	67.85	11.13	114.2	1.52
	135P-400K	low	263.0	3240	168.7	2378	258.7	3176	12.52	166.5	9.33	117.8	9.56	109.2	1.56

Statistical analysis were carried out using the analysis of variance according to the factorial design (2 x 2 x 3) and eventually combined with the new multiple range test of DUNCAN (1955).

Two types of results can be considered :

1. In case of no interaction between the factors studied the results were formulated in terms of main effects,

   I. e. Mg (- & +)

   N (low and high)

   PK (PK_1, PK_2 and PK_3)

2. When interaction is present, one factor is studied within the interacting one, I. e. if N (pK) is significant, the effect of N is considered within each PK treatment and vice versa.

Results of the analysis :

Table 46. 1

Type I T. 67. 10 main effect		
Element	Treatment	Statistical results
Cu	- Mg + Mg	oo
	HN Low N	oo
	(PK_1) (PK_2) = (PK_3)	oo
Pb	- Mg + Mg	oo
	High N Low N	o
	(PK_1) (PK_2) = (PK_3)	oo
Fe	- Mg + Mg	oo
	(PK_1) (PK_2) = (PK_3)	oo

Table 46.2

Type II T.67.10 Interaction		
Element	Treatment	Statistical results
Zn	$- Mg < + Mg$	**
	Effect of PK treatment within HN :	
	$(PK_1) > (PK_2) = (PK_3)$	**
	Effect of PK treatment within LN :	
	$(PK_1) = (PK_2) = (PK_3)$	-
	Effect of N within (PK_1):	
	High N > Low N	**
	(PK_2) HN = LN	
	(PK_3) HN = LN	

Table 46.3

Type I T.67.12 Main effect		
Element	Treatment	Statistical results
Mn	$- Mg > + Mg$	*
	High N > Low N	**
	$(PK_1) > (PK_2) > (PK_3)$	**
Cu	High N > Low N	**
	$(PK_1) > (PK_2) > (PK_3)$	**
B	High N < Low N	*
	$(PK_1) > (PK_2) = (PK_3)$	**
Pb	High N > Low N	**
	$(PK_1) > (PK_2) > (PK_3)$	**
Ni	High N > Low N	*
	$(PK_1) > (PK_2) = (PK_3)$	**

Table 46.4

Type II T.67.12 Interaction		
Element	Treatment	Statistical results
Fe	Effect of Mg treatment within (PK_1) : + Mg > - Mg	**
	Effect of Mg treatment within (PK_2) : + Mg = - Mg	-
	Effect of Mg treatment within (PK_3) : + Mg = - Mg	-
	Effect of PK within - Mg : $(PK_1) > (PK_2) > (PK_3)$:	*
	Effect of PK_3 within + Mg : $(PK_1) > (PK_2) > (PK_3)$:	**
	Effect of N within (PK_1) : higher N > lower N	**
	Effect of N within (PK_2) : higher N > lower N	**
	Effect of N within (PK_3) : higher N ≑ lower N	-
	Effect of PK within higher N: $(PK_1) > (PK_2) = (PK_3)$	**
	Effect of PK within lower N: $(PK_1) > (PK_2) = (PK_3)$	**
Zn	Effect of Mg within higher N : + Mg = - Mg	**
	Effect of Mg within lower N : + Mg > - Mg	**
	Effect of N within - Mg : Higher N > lower N	**
	Effect of N within + Mg : higher N > lower N	*
	Effect of PK within lower N: $(PK_1) > (PK_3) = (PK_2)$	*
	Effect of PK within higher N:	
	$(PK_1) > (PK_3)$	**
	$(PK_2) > (PK_3)$	**
	$(PK_1) \doteq (PK_2)$	-
	Effect of N within (PK_1) : higher N = lower N	
	Effect of N within (PK_2) : higher N > lower N	**
	Effect of N within (PK_3) : higher N > lower N	**

* significant at 5 % ; ** significant at 1 %

4. CONCLUSIONS

The trace element absorption by pasture plants as studied in this part, shows to be also subject to variations in function of short term differences in major element fertilization. The differences in trace element contents were mainly due to high and low phosphorus and potassium treatments. In the special case of manganese less variation was observed, due to the fact that the former treatments did not cause substantial pH variations in the short time experiment.

CHAPTER VI

PRELIMENARY OBSERVATIONS CONCERNING METAL CONTAMINATION IN BELGIAN GRASSLAND

A. AN EVALUATION OF THE EXTEND OF HEAVY METAL CONTAMINATION NEAR MAJOR HIGHWAYS, INDUSTRIAL AND URBAN AREAS.

1. INTRODUCTION

An investigation was made on zinc, copper and lead contents in soils and grass samples collected along highways and field adjoining industrial zones. The problem of air pollution and contamination due to automobile exhausts in busy highways and in important industrial areas appears to be rather important.

Recently, investigations from different countries have been reported. WARREN and DELVAULT (1960, 1962), found that plants subjected to atmospheric pollution or grown in soils high in lead may contain 10 times the normal amounts of that element. They showed that it originated from the exhaust fumes of motor vehicles using leaded petrol.

Contamination with lead from car exhausts originating in tetra ethyl lead in petrol is well established phenomenon. This has been confirmed by CANNON and BOWLES (1962) and others.

Under different nutrient conditions (Chapter V) the lead content of herbage plants was generally found in the range between 7 - 12 ppm, while by late winter, the amount increased to 25 - 33 ppm Pb. In soils, lead content was found to vary over a large range, as well in total as in extractable values [MITCHELL and REITH (1966)]. In eastern Canada, surface samples of 10 cultivated soils contained 6 to 14 ppm lead (unpublished data, Canada Dept. of Agr. Ottawa). WRIGHT et al. (1955) reported cases of a pronounced accumulation of lead in A_O horizon of podzolic soils.

One sample contained 108 ppm. In Scotland, SWAINE and MITCHELL (1960) have pointed out that surface horizons of most soils are often considerably richer in lead than the lower horizons. They reported an instance of 550 ppm lead in the A_1 horizon of a Scottisch soil. In a comparison of 19 orchard and other soil samples in Nova Scotia, CHISHOLM and BISHOP (1967) found that the lead content was usually higher than 50 ppm in the former, and below this amount in the latter ; the highest value was 360 ppm. Urban garden soils were found by PURVIS (1967) to contain 20.4 ppm.

There is no evidence that the lead content of contaminated soils is directly proportional to the amounts taken up by plants. MARTEN and HAMMOND (1966) found only 2.5 ppm lead in the first crop of bromegrass grown in a contaminated sandy loam soil containing 680 ppm. They cite the work of KLOKE and RIEBARTSCH (1964), who concluded that grass accumulated considerable amounts of lead in the roots, but that only limited quantities were translocated to the above-ground parts. In Western Ireland, the hazard of lead intake by workers engaged in lead mining operations and industrial occupations, was reported by DONOVAN et al. (1968) (1969).

The same authors drew the attention to :

1. the dust fallout from the mining operations on the adjoining fields and its possible direct or indirect hazard to animal life,
2. the need to test food supplies in the area,
3. and the danger of contamination of milk and butter from cows grazing on such pastures.

MACLEAN et al. (1969) reported that the lead concentration in different plant species increased with decreasing distance from a busy highway.

Other contaminating trace elements may originate from different sources. PURVIS (1966, 1967, 1968 and 1969) confirmed that soils in urban parklands are often contaminated with copper, boron, lead and zinc.

Herbage from the same areas contained significantly higher levels of lead and zinc than herbage in rural areas.

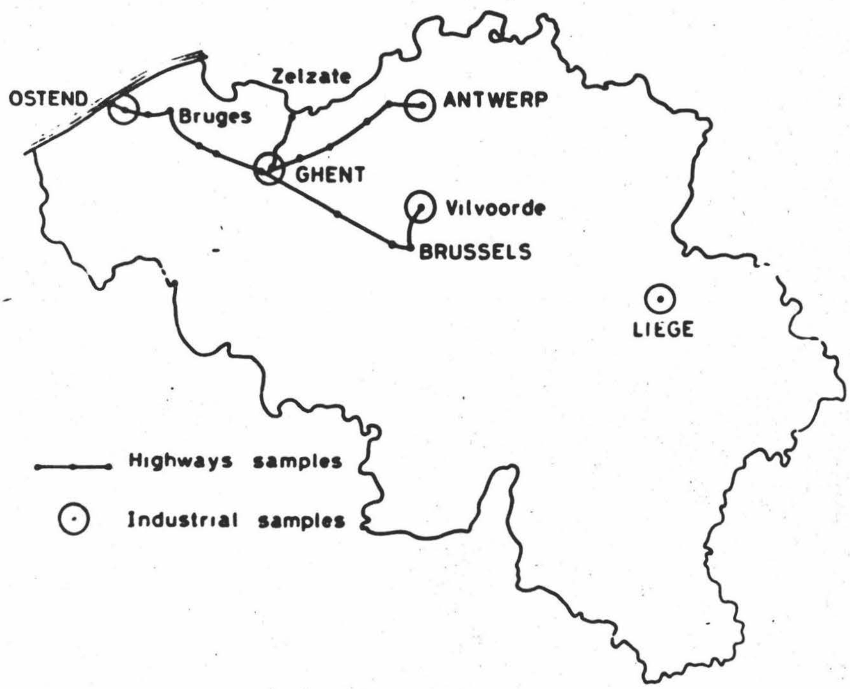
The object of the first part of the present study was :

1. to evaluate the contamination of soils and plants by zinc, copper and lead along some major highways in Belgium.
2. to study the rate of contribution of dust "Fallout" from industrial areas on pasture crops adjoining them.

2. MATERIAL AND METHODS

Soils and herbage plants were sampled along the highways : Brussels - Ostend and Antwerp - Ghent. The samples were collected during the years 1968-1969, from cultivated and uncultivated pasture lands in proximity to the highway.
Other samples were taken from fields in the following industrial regions : Liège, Antwerp, Vilvoorde, Ghent and Ostend (fig. 24).

FIG. 24 PLANT AND SOIL SAMPLES COLLECTED ALONG HIGHWAYS AND INDUSTRIAL AREAS IN BELGIUM



The herbage samples included grass and clover species. The plants were clipped about 5 cm above ground level and were not washed before trace element analysis. The soil samples were air dried, crushed and extracted with O. 5 N nitric acid for the trace element determination.
Trace elements in soil and plant were determined by direct reading emission spectrography (COTTENIE e. a. 1966-1967).

3. RESULTS AND DISCUSSION

3. 1. Trace element contamination of soils near highways

Our analysis show that soil contamination with zinc and lead is a quite pronounced phenomenon along the highways and in some cases, contamination with manganese was also observed.
This clearly appears when one compares the results given in tables 47. 1 & 47. 2 showing the range of trace elements normally encountered in different agricultural regions of Belgium. Furthermore this contamination seems to decrease rapidly with increasing distance from the highway.
In some cases the soil analysis indicated a higher amount of lead and manganese at a distance of 8 m than just beside the road, but all our results show that the soil contamination remains a local one, which is not extended to distances of 2O m or more. These results are in accordance with those obtained by CANNON and BOWLES (1962) and others.
There was no indication of real contamination with other elements for instance with copper.

3. 2. It also seemed interesting to analyse soil samples from industrial areas in order to evaluate the presence of trace elements and to observe any eventual contamination. This was made in different regions and the analytical results are given in tables 47. 3 and 47. 4. These results show that generally the elements zinc and lead are present and much higher levels than the normal ones, and that in some cases there is also an enrichment in Mn, Cu, Ni and Co.

3. 3. Trace elements contamination in urban gardens

In Scotland PURVIS (1968) found a remarkable enrichment of urban garden soils with different trace elements : EDTA extractable copper was five times higher than the normal content of that element in rural soils, water soluble boron twice, acetic acid extractable lead and zinc respectively 5 and 25 higher than normal. Analysing the "root" of CHEMNIS he found that this product contained 555 to 64O ppm water soluble B.

Other sources of contamination might be in these conditions domestic coal ash and municipal compost.

Table 47. 5 shows an example of the trace element concentrations found in two small urban gardens (Ghent and Brussels), compared with the mean normal contents and with the values advanced by PURVIS (1967 - 1968). The reported val ues from Ghent and Brussels illustrate also the existance of at least Zn, Pb and Cu contamination.

Table 47.1 : Extractable (0.5 n HNO_3) trace elements in soils collected near the highway Brussels - Ostend (13.6.69 and 20.6.69) - in ppm.

Location and proximity to the highway	Al	Fe	Mn	Zn	Cu	Pb	Ni	Si	Mo	Co	Cr	pH H_2O	pH KCl
Groot-Bijgaarden	688	963	112.5	>55	6.75	>55	2.5	700	traces	4	traces	6.59	6.12
Aalst													
old part 12 m	388	220	50.0	10.0	2.50	>55	traces	50	traces	traces	traces	5.63	4.53
new part 3 m	538	435	144.0	9.3	2.75	15.5	traces	463	traces	traces	traces	7.65	7.11
8 m	1085	530	225.0	12.0	3.50	16.0	traces	1463	traces	traces	1.0	7.79	7.20
20 m	705	325	26.5	5.0	1.50	2.5	traces	260	traces	traces	traces	6.35	5.02
Drongen 1 m	>1200	513	177.5	>55	8.50	>55	2.0	>1500	traces	1.0	2.0	7.40	7.02
5 m	425	425	56.5	24.5	3.50	25.8	traces	275	traces	traces	traces	7.05	6.66
Aalter 1 m	>1200	400	205.0	>55	6.50	>55	2.0	>1500	traces	traces	2.0	7.09	6.68
Beernem 1 m	1075	380	110.0	38.0	3.50	>55	traces	133	traces	traces	1.0	6.80	6.12
Brugge 1 m	>1200	470	>300	>55	3.50	>55	1.0	>1500	3.0	traces	3.5	7.12	6.73
Jabbeke 1 m	763	563	92.5	25.8	4.50	>55	traces	875	traces	traces	0.5	6.89	6.49
St. Michiels 1 m	>1200	338	182.5	29.5	16.0	>55	traces	± 1500	traces	traces	traces	7.07	6.79
Mean normal content ±	550	500	65	11	6.0	11	traces	-	traces	traces	traces	-	-

Table 47.2 : Extractable (0.5 n HNO_3 extraction) trace elements in soils collected near the highway Antwerp-Ghent (15.6.69) in ppm.

Location and proximity to the highway		Al	Fe	Mn	Zn	Cu	Pb	Ni	Si	Mo	Co	Cr	pH	
													H_2O	KCl
Antwerp	5 m	600	713	81.5	>55	23.5	>55	traces	688	traces	traces	0.5	7.35	7.20
Beveren-Waas	0.5 m	1200	675	144.0	>55	11.75	>55	traces	>1500	traces	traces	0.5	6.45	6.02
	10 m	615	500	5.0	4.3	3.75	14.0	traces	traces	traces	traces	traces	4.92	4.15
St. Niklaas	0.5 m	390	313	63.5	9.5	2.0	10.5	2.0	30	traces	traces	traces	6.59	5.92
new part	1 m	575	825	97.5	9.0	8.0	13.5	traces	traces	traces	traces	0.5	6.04	5.20
Lokeren	0.5 m	850	575	86.5	>55	13.25	>55	traces	1070	traces	traces	2.25	7.09	7.01
	10 m	>1200	338	85.0	20.5	6.0	25.0	traces	138	traces	traces	0.5	5.89	5.10
Lochristi	0.5 m	>1200	500	194.0	>55	14.5	>55	traces	>1500	traces	traces	2.25	7.12	6.79
	10 m	838	788	40.0	14.0	10.3	30.0	traces	traces	traces	traces	0.5	5.59	4.78
Mean normal content ±		550	500	65	11	6.0	11	traces	-	traces	traces	traces	-	-

Table 47.3 : Extractable trace element contents in soils from industrial areas (Brabant and Antwerp) ppm (0.5 n HNO_3 extraction)

	Willebroek				Vilvoorde		Antwerp	Mean normal content (order of magnitude)
	a grassland near factories	b grassland near factories	c grassland near factories	d 100 - 200 m distance	a (Industrial area)	b	(Industrial area	
Al	600	390	538	630	>1200	>1200	263	500
Fe	>1200	763	1010	580	1100	525	488	400
Mn	114	57.5	130	55	127.5	>300	72.5	70
Zn	>55	42	>55	>55	>55	>55	15.0	10
Cu	11	6.7	145	15	>55	19.5	2.25	7
Pb	>55	45	42.3	55	>55	>55	7.00	19
Ni	2.7	traces	1.5	traces	4	1.5	3.00	-
Mo	traces	traces	traces	traces	3.0	traces	12.00	traces
Co	traces	traces	1.5	traces	2.0	1.75	2.5	-
Cr	traces	traces	0.5	traces	1.0	1.0	0.5	-
Si	traces	traces	525	88	728	>1500	188	-
pH-H_2O	5.53	4.60	7.26	4.99	6.30	7.50	7.82	
pH-KCl	4.78	3.68	6.97	4.08	5.89	7.20	7.50	
	13.5.69	13.6.69	2.7.69	2.7.69	2.7.69	2.7.69	15.7.69	

Table 47.4 : Extractable trace element contents in soils from industrial areas (Gent and Tertre)

in ppm (0.5 n HNO_3 extraction)

	Desteldonck	Zelzate	Terdonck	Tertre at 100 m	Tertre at 500 m	Normal content
Al	755	605	1088	520	395	500
Fe	938	>1000	863	463	263	400
Mn	128	143	137	288	43.5	70
Zn	11.2	16.8	16.0	25.8	12.8	10
Cu	6.2	8.8	3.8	3.0	2.5	7
Pb	24	40	38	traces	traces	12
Ni	traces	traces	traces	11.0	14.3	
Mo	traces	traces	traces			
Co	traces	traces	traces			
Si	37.5	55	113	50	<25	
pH-H_2O	5.4	6.9	5.8	6.9	5.9	
pH-KCl	5.0	6.2	5.0	6.4	4.9	

Table 47.5 : Extractable trace element in soils from urban gardens - in ppm (0.5 n HNO_3 extraction)

	Ghent	Brussels	Mean normal contents	Mean values PURVES D. Urban gardens	(Scotland) Normal content
Al	>1500	1200	550	-	-
Fe	910	>538	500	-	-
Mn	24	>300	65	-	-
Zn	>25	>55	11	52.4	2.7 (a)
Cu	48	42	6	T 66.8 E 16.7	15.5 (b) 2.8 (c)
B	-	-	-	1.87	0.70(d)
Pb	>55	>55	11	2.89	0.65(e)
Mo	-	4			
Co	-	5.8			
$pH-H_2O$	4.9	6.8			
pH-KCl	3.7	6.1			

T total
E extractable

a Zn soluble in acetic acid
b Total Cu
c EDTA extractable Cu
d water soluble B
e Pb soluble in acetic acid

Table 47.6 : Trace element contents in soils from Sart-Timan (Liège) in ppm (Total T and 0.5 n HNO_3 extraction E)

	A		B		C		Mean normal content E
	T.	E.	T.	E.	T.	E.	
Al	30000	825	29000	750	28500	795	500
Fe	25600	395	24000	350	28400	350	400
Mn	730	186	640	155	570	200	70
Zn	2725	1400	2725	1400	3850	1600	10
Cu	82	39	87	42	122	50	7
Pb	230	230	260	260	230	240	12
Ni	72	3.5	60	2.5	82	3.5	2
Mo	traces	traces	traces	traces	25	traces	1
Co	42	traces	40	traces	50	traces	1
B	68	-	60	-	68	-	-
pH-H_2O	7.9		8.0		7.8		
pH-KCl	7.2		7.2		7.0		

T. total

E. extractable

4. TRACE ELEMENT CONTENTS IN PLANTS GROWING ON CONTAMINATED SOILS

As has been shown in this work, the trace element content of plants is largely variable in function of soil conditions. However there is no evidence to state that high amounts in the soil would a priori indicate similar high levels in the plants. Indeed the mobility and availability of the trace elements is governed by several factors such as pH and organic matter content.

In order to estimate the contamination of plants growing in the areas earlier mentioned, grass samples were taken on the same places. Tables 48. 1 and 48. 2 indicate that the main contamination is an increased content of lead and only occasionally of some other trace elements such as Mn and Zn. This is confirmed by CANNON and BOWLES (1962) who also found increased lead content in the same conditions. This contamination however decreased considerably with the distance from highways.

It seems difficult to distinguish between the amounts of lead taken up from the soil by the roots and the direct contamination of the leaves. In connection with this point MARTEN and HAMMOND (1966) found only 2. 5 ppm lead in bromegrass grown in a contaminated sandy loam containing 68O ppm Pb. They cite the work of KLOKE and RIEBARTSCH (1964) who stated that considerable amounts of lead taken up by the roots were not necessarily translocated to the upper parts of the plants.

Concerning the other trace elements it seems not very likely that elements such Fe and Mn could be much affected because their mobility is strongly pH dependent. There is on the other hand a much greater possibility for increased absorption of Zn, Cu and B, the mobility of these elements being less pH dependent. Also the non essential trace elements Pb, Ni, Cr etc. might be taken up by the plants in more than normal contents when they should be present as contaminants (PURVIS 197O).

One of the more specific contaminations concerns fluorine, which can be brought into the soil and the plants by fallout in some industrial areas. This kind of contamination was observed by WEBSTER (1967) and OELSCHLAGER (1968). In Belgium VERLOO and COTTENIE (1970) found contaminated grass containing up to 200 ppm F in comparison with a normal content of less than 10 ppm.

Table 48.1 : Trace element content of herbage plants, sampled near the highway Brussels - Ostend (ppm in dry matter)

	Distance from road (m)	Al	Fe	Mn	Zn	Cu	Pb	Ni	Mo	Co	Cr
Groot-Bijgaarden	1	448	870	95	66.7	21.6	>100	6.35	1.52	2.54	5.08
Groot-Bijgaarden	1	321	688	73	63.5	26.2	>100	5.64	2.26	3.05	6.49
Groot-Bijgaarden	5	230	335	87	31.3	11.1	9.6	3.11	0.84	0.84	2.87
Aalst - old part	12	101	185	343	38.4	12.2	6.1	2.26	traces	traces	2.26
new part	3	393	470	71	38.4	16.2	9.7	4.14	2.37	2.96	2.48
	8	164	266	67	40.2	12.6	17.5	1.12	0.42	traces	1.26
	20	305	396	119	35.0	15.8	15.0	6.09	2.44	4.63	3.65
Drongen	1	395	720	64	114.2	27.2	>100	10.54	6.67	7.61	2.81
Drongen	5	92	208	28	48.8	14.7	18.9	traces	traces	traces	0.31
Aalter	1	192	406	52	90.4	18.4	>100	4.06	1.32	1.10	1.37
Beernem	1	188	361	61	100.3	18.8	>100	4.06	2.03	1.73	1.27
Brugge	1	133	262	39	67.5	14.1	>100	0.48	0.29	traces	0.73
Glabbeke	1	189	347	56	79.1	14.8	39.8	1.73	traces	traces	1.28
St. Michiels	1	95	169	25	37.1	10.1	30.1	2.06	0.83	traces	0.50
Normal contents		145	236	55	61	7.5	2.1	traces	traces	traces	traces

Table 48.2 : Trace element content of herbage plants near the highway Antwerp-Ghent (ppm in dry matter)

	Distance from road (m)	Al	Fe	Mn	Zn	Cu	Pb	Ni	Mo	Co	Cr
Antwerp	0,5	279	603	59	78.8	17.6	42.8	5.40	3.42	1.36	1.80
"	5,0	40	126	86	43.0	12.6	12.6	traces	traces	traces	traces
Beveren Waas	0,5	>800	>1000	129	134.1	42.1	42.1	11.55	2.72	3.13	9.17
"	10,0	79	420	>600	65.0	15.1	19.1	3.63	1.82	0.85	0.54
St. Niklaas	0,5	582	>1000	144	105.8	26.1	>100	8.23	1.41	2.0	3.53
"	10,0	63	161	137	37.0	10.0	14.3	traces	traces	traces	traces
Lokeren	0,5	>800	>1000	130	139.4	31.0	>100	10.53	3.10	4.34	5.45
"	10,0	69	174	108	58.4	11.4	10.8	traces	traces	traces	traces
Lochristi	0,5	739	>1000	169	169.0	36.6	>100	10.56	traces	1.41	5.49
"	10,0	140	319	200	71.1	15.8	20.9	5.34	0.86	1.21	0.86
Normal contents		145	236	55	61	7.5	2.1	traces	traces	traces	traces

B. MOBILITY OF METAL IONS IN CONTAMINATED SOIL AS AFFECTED BY APPLICATIONS OF LIME AND CHELATING AGENTS, WITH REFERENCE TO PASTURE CROPS.

1. INTRODUCTION

Since hazard distributions of metallic ions in soils and plants is increasingly reported, it is important to study mechanisms of possible immobilization of these elements. The most important factors which influence the mobility of trace elements in the soil are the pH and the organic matter. In the preceeding part both these factors have already been mentioned in connection with trace element uptake.

pH changes are most often realized by addition of lime and sulfur. The addition of lime decreases the availability of most micronutrients while sulfur acts in an opposite way. (BROWN et al. (1964): (CHRISTIANSEN et al. 195O), (FERGUS 1954, 1956), (SANCHEZ et al. 1959), (SEATZ et al. 1959), (SHERMAN et al. 1941) and (WEAR 1956).
The organic matter can interact with trace elements in different ways. Its role in connection with ion adsorption and exchange has been recognized already in the early stage of chemical soil studies. The formation of organo-metallic complexes was increasingly studied during the last decade.
The behaviour of natural soil humic compounds was compared with synthetic chelating agents and the effect of the latter on the availability of trace elements such as Fe, Zn, Cu and other appears to be a promising field of soil improvement. Characteristics of chelating agents were reported by HODGSON (1968), WALLACE et al. (1956) and WALLACE et al. (1968).
HODGSON (1967) suggested that complexing agents may increase the availability of trace elements by increasing their participation in the mass flow to the roots through the soil.
In fact one must distinguish between two types of complexes soluble and insoluble ones. The former will contribute to a higher

availability if the plant roots are capable to take up such large molecules. The latter will join the solid phase to the soil and represent a form of immobilisation.

Each one of these cases may be of interest, namely in connection with eventual deficiencies or in immobilising toxic amounts of trace elements.

All trace elements are toxic to both plants and animals if present in high concentrations, but some species are more sensitive than others. As an increasing number of cases of trace element accumulation in the soil up to the toxic level was reported.

Differences between various crop plants in their resistance to injury have been reported by VERGANO (1953), BRADSHAW (1952), JOWETT (1956), SCHMEHL et al. (1950), HALLSWORTH et al. (1957), DESSUREAUX and OUELETT (1958), TURCIN and SOKOLOV (1950).

Two pot experiments were set up in order to study the mobility and uptake of trace elements by plants under the influence of pH changes and presence of chelating agents.

2. FIRST POT EXPERIMENT

2.1. Experimental details :

The experiment was started in December 1968 using a soil contaminated with Zn, Cu and Pb originating from an industrial area in the "KEMPEN". Plastic pots containing 1 kg of soil, kept at field moist capacity with deionized water, were treated with the following combination of lime (CaO) and Na_2EDTA :

lime as kg CaO per ha	Na_2EDTA in g per kg of soil
2500	0
	0.5
	1.0
5000	0
	0.5
	1.0

After treatment the pots were incubated for 14 days. Initially Agrostis Tenuis was sown as an experimental crop but these plants did not resist and on the 15. 1. 1969 75 seeds of Perennial Ryegrass (R. v. P.) were resown. All the treatments were repeated four times and the layout was in the form of a randomized bloc.

The soil characteristics were determined as follows :

pH-H_2O	5. 40
pH-KCl	4. 65
C. E. C.	3. 20 meq per 100 g

The total trace element contents, ppm in air dry sample, as determined by direct reading spectrography, were :

Al	2400	Cu	40
Fe	2850	Pb	110
Mn	35	Co	10
Zn	630	Ni	18

After the first two weeks the pH increased up to 6. 45 in the pots with 2500 kg CaO/ha and to 7. 15 in the pots with 5000 kg CaO/ ha. Since it was not possible to grow plants on the original non-limed soil, the experiment does not contain a control without lime.

2. 2. Results

The plants provided one harvest after two months of growth, but were showing toxicity phenomena in varying degrees so that it was not possible to grow more cuttings. The dry matter yields and percent of ash, as affected by the tre atments, are shown in table 49.

Table 49 : Dry matter yields and ash content

Treatment	Soil pH	mean dry matter yield per pot (g)	% ash
CaO (2500)	6.45	0.232	9.97
+ Na_2EDTA (1)	6.45	0.267	15.26
+ Na_2EDTA (2)	6.45	0.059	19.32
CaO (5000)	7.15	0.337	15.76
+ Na_2EDTA (1)	7.15	0.489	16.23
+ Na_2EDTA (2)	7.15	0.195	18.00

The trace element concentrations in the plant tissues and the total uptake are given in tables 50 & 51.

2.3. Discussion

2.3.1. Yield : As a first observation the yields were systematically increased by the highest rate of lime, which raised the pH up to 7.15. Within each lime treatment, the application of EDTA also had a significant influence. With 0.5 of Na_2EDTA/kg of soil a yield increase was observed, while an important decrease was caused by 1 g Na_2EDTA per kg of soil.

2.3.2. Iron and manganese :

The absorption of iron varied largely with the different treatments and the influence of EDTA at pH 6.45 was most pronounced. After the addition of 1 g EDTA per kg of soil, the iron content of the plants was three times the value obtained without EDTA. This is in accordance with LACHICA et al. (1967) who stated that the concentration of iron in the plant increases with the quantity of chelating agent added.

Once again manganese showed a clear antagonism with iron and this resulted even in a higher manganese uptake at higher pH. This is in contradiction with the normal manganese response towards pH variations. While EDTA positively influenced the iron concentrations in the tissues, the contrary influence was observed with regard to manganese. The total uptake of both elements did not follow the same trend as their concentrations, due to the large differences in dry matter yields.

2.3.3. Zinc :

As the soil being used was highly contaminated with zinc the concentrations of this element were generally very high.

The zinc conte nt of the shoots was also strongly influenced by EDTA, the highest values being found with 0.5 g EDTA at pH 6.45 and with 1 g EDTA at pH 7.15. This is an indication for the fact that more EDTA is involved in the mobilisation of zinc at higher pH values.

Concerning the total uptake of this element, the same effect however was more or less levelled off by the differences in dry matter yield.

2.3.4. Copper :

Even the lowest copper contents being observed on this contaminated soil are still to be considered as very high. At each pH level the applications of EDTA gave a significant increase of copper concentration and this effect was more pronounced at pH 7.15 than at pH 6.45 (table 50).

In spite of these differences in copper content, no significant influence on the total copper uptake was observed. This means that the increase of Cu concentration as an effect of EDTA, completely compensated the yield decrease earlier mentioned.

2.3.5. Boron :

All the plants were characterised by low boron contents, and the tissue concentrations of this elements decreased significantly with increasing rates of EDTA (table 50). This might be an indirect effect of the treatments and be attributed mainly to a phenomenon of antagonism.

The boron contents obtained in the plants of the EDTA treated pots range within the level of deficiency. Indeed critical values reported by different authors are from 6.9 to 23.0 ppm, the mean critical level being considered as near 9 ppm in clover (ROGERS 1947, DIBLE & BERGER 1952). Due to the very pronounced influence of EDTA on the boron content, the total uptake of this element follows the same pattern, in spite of the yield fluctuation.

Remark : As table 50 also contains the analytical results for nickel, it seems intresting to mention that this element behaved in a very similar manner as boron.

2.3.6. Lead :

As the soil was highly contaminated with lead the plant tissue contents were very high in lead, and the analysis revealed figures up to 382 ppm, whilte the normal content is ± 10 ppm.

With respect to this element, there was a similar effect as for zinc. This means that both lime and EDTA had an influence on its uptake. The highest values were obtained with 0.5 g EDTA at the first lime level (pH 6.45) and with 1.0 g EDTA at the second lime level (pH 7.15). In any case EDTA had a positive influence on the uptake of lead.

Table 50 : Effect of lime and chelating agent applications on the trace element concentration (ppm in dry matter)

Soil pH	lime (CaO) kg/ha	Na_2EDTA g/kg soil	Concentration ppm in dry matter						
			Fe	Mn	Zn	Cu	B	Pb	Ni
6.45	2500	0	129	179	770	25.92	6.89	18.17	13.52
		0.5	303	100	2077	44.57	1.72	158.2	4.60
		1.0	372	102	1327	48.20	traces	118.1	traces
		Linear regression	**	**	*	*	**	N.S.	*
7.15	5000	0	155	194	856	31.30	6.94	50.70	12.90
		0.5	101	164	984	27.52	1.88	125.7	4.37
		1.0	163	174	2266	59.47	0.38	328.2	0.47
		Linear regression	N.S.	N.S.	**	**	**	**	**

* - significant at 5 %

** - significant at 1 %

N.S. - not significant

Table 51 : Effect of lime and chelating agent applications on the trace element uptake (µg/pot)

			uptake - µg/pot						
Soil pH	lime (CaO) kg/ha	Na_2EDTA g/kg soil	Fe	Mn	Zn	Cu	B	Pb	Ni
6.45	2500	0	30.35	42.13	186.5	6.12	1.68	4.22	3.23
		0.5	80.47	26.73	547.3	11.93	0.46	41.12	1.28
		1.0	40.10	11.47	155.0	5.19	traces	13.45	traces
		Linear regression	N.S.	**	N.S.	N.S.	**	N.S.	*
7.15	5000	0	49.54	63.81	317.3	10.16	2.02	6.64	3.44
		0.5	46.14	76.40	501.3	12.89	0.54	68.26	1.25
		1.0	31.18	33.78	446.0	11.25	0.06	73.46	0.80
		Linear regression	N.S.	**	N.S.	N.S.	**	**	*

* - significant at 5 %

** - significant at 1 %

N.S. - not significant

3. SECOND POT EXPERIMENT (Zn-contaminated soil)

3.1. Introduction

In 1969 a soil highly contaminated with zinc from an industrial region gave the analytical results shown in table 52 (Trace elements extractable with 0.5 n HNO_3).

				$pH-H_2O$	pH-KCl
Fe	638	Co	1.50	7.25	6.50
Zn	3030	Ni	1.75		
Al	463	Cr	5.0		
Mn	60	Si	2.05		
Cu	7.75	Mo	traces		
Pb	44.5				

The zinc content of this soil was several hundred times higher than the normal mean content of Belgian soils, which is 11 ppm.
Oat plants grown on this soil developed much more slowly, than in a normal soil and had a leaf content of 1955 ppm Zn in comparison with 62 ppm in a normal soil.
A pot experiment was carried out, in which treatments with lime, peat and DTPA (Diethylene triamine penta-acetic acid) were applied in order to observe the influence of these products on the zinc behaviour in the soil and its uptake by the plants. The general scheme of the experiment, in which barley and Italian ryegrass were used as test crops, is given as follows :

DTPA mg per kg soil	Peat % added to the soil	lime as kg CaO per ha	Control
A 250	C 5 %	E 2000	G
B 500	D 10 %	F 4000	

The pots contained 1 kg of soil and all the treatments were carried out with four replications. At the same time the whole experiment was replicated with normal soil as a reference.

3.2. Results

3.2.1. Soil acidity

After the experiment the pH H_2O and pH KCl were measured and this gave the results shown in table 53. From this table it appears that the peat addition as well as the DTPA treatment have significantly lowered the soil pH.

Table 53 : Soil pH after pot experiment no 2.

	Zn-contaminated soil		Reference soil	
Treatment	pH-H_2O	pH-KCl	pH-H_2O	pH-KCl
DTPA A - 250	6.59	5.99	6.00	4.74
B - 500	6.00	5.94	5.95	5.74
Peat C - 5 %	6.16	5.67	5.24	4.17
D - 10 %	5.74	5.28	4.80	3.91
Lime E - 2000	6.91	6.30	7.05	6.01
F - 4000	7.56	6.88	7.63	6.84
Control G	6.49	5.95	5.93	4.68

3.2.2. Dry matter yield

The dry matter yields for both experimental crops are given in table 54.

Table 54 : Dry weight of shoot of barley and Italian ryegrass as influenced by different treatments.

	Barley		Italian ryegrass	
	Zn-contaminated soil	reference soil	Zn-contaminated soil	reference soil
Treatment	g	g	g	g
DTPA A - 250	1.66	1.96	0.33	2.08
B - 500	1.68	2.05	0.30	2.09
Peat C - 5 %	1.18	1.90	0.37	2.31
D - 10 %	1.19	1.82	0.29	2.37
Lime E - 2000	1.95	2.22	0.74	1.89
F - 4000	2.21	2.09	1.34	2.34
Control G	1.39	2.22	0.27	2.22

a) in the case of barley all the treatments, except the ones with peat, increased very significantly the yields in comparison to the non treated zinc enriched soil (control).
The most favourable treatment appeared to be the application of lime, while also DTPA had a very positive influence with regard to the dry matter yield.

b) Italian ryegrass reacted positively only towards lime, giving dry matter yields, which were four times higher after the highest liming rate than in any other case.
The Italian ryegrass gave a second cutting in the case with the highest liming, while in the other pots, the growth of the plants almost stopped completely.

3.2.3. Trace element absorption

The trace elements Fe, Mn, Zn and Cu were determined in the plants by atomic absorption.

The mean results of these analysis are given in tables 55 & 56.

While the zinc contents, obtained on the reference soil, ranged between 40 and 75 ppm in barley leaves, and between 58 and 117 ppm in ryegrass, the plants grown on the zinc contaminated soil showed extremely high zinc contents ranging till 4155 ppm in barley and till 18000 ppm in ryegrass.

These high concentrations in the leaves were only affected significantly by the lime treatments :

- in barley, liming reduced the zinc content till 1575 ppm and 887 ppm respectively, after the addition of 2000 and 4000 kg of CaO/ha. Comparing these values with the control, in which the zinc concentration was 3606 ppm, this means a reduction of zinc absorption with a factor 2 to 4.
- In ryegrass, the two lime levels resulted in zinc contents of respectively 2875 and 950 ppm, which corresponds in comparison with the 16300 ppm of the control, to a reduction by a factor of approximatively 5.5 and 10 respectively. In this case the peat addition also reduced somewhat the concentration.

In spite of the favourable effect of liming, all the plants grown on the Zn contaminated soil, were still very high in Zn content in comparison to the plants of the reference experiment.

On the reference soil DTPA and peat stimulated the uptake of manganese, but there was no other clear cut effect of the treatments with regard to the trace element concentration in the plants.

C. CONCLUSION

Observations in the field, as well as in pot experiments, show the high sensitivity with which pasture crops react towards mineral contamination of the soil. This affects as well the growth and dry matter production as the mineral composition and the quality of the plants. High increase of trace element contents in the tissues occurs more easily on acid soils, where the mobility of the elements is the highest.

In all cases, raising the pH of the soil by liming, appeared to be the most important factor for reducing the absorption of toxic levels of trace elements. However the mobility of Fe and Mn is much more pH dependent than the zinc, copper and boron. In the case of very high contamination levels, the usual liming rates, bringing the soil to neutrality, seem still to be insufficient for reducing the trace element absorption to the normal values.

Complexing agents generally act in an opposite way, their effect being normally more pronounced in less acid conditions.

As far as our observations reach, trace element contamination of soils is occuring locally in relatively small industrial areas and along the main roads.

Carefull and systematic observation of this extention is however to be recommended, where industrial developement continuous to occupy more and more rural sites.

Table 55 : Trace element concentrations (p.p.m. in dry matter) in barley shoots under various treatments.

	Zn-contaminated soil				Reference soil			
	ppm in dry matter				ppm in dry matter			
Treatments	Fe	Mn	Zn	Cu	Fe	Mn	Zn	Cu
DTPA A - 250	60.6	51.8	3447	11.1	124.0	51.9	75.4	13.4
B - 500	90.1	73.8	4115	14.6	88.9	42.9	49.7	10.5
Peat C - 5 %	67.5	131.6	3512	13.9	69.2	171.2	56.7	13.6
D -10 %	80.8	148.8	3784	18.7	52.4	255.4	57.9	11.8
Lime E - 2000	90.0	91.9	1575	11.2	55.1	59.7	50.2	9.2
F - 4000	74.9	97.6	887	10.5	66.7	81.0	65.4	10.7
Control G	98.0	82.3	3606	16.2	62.4	41.1	46.1	11.3

Table 56 : Trace element concentrations (p.p.m. in dry matter) in Italian ryegrass under various treatments.

	Zn-contaminated soil, ppm in dry matter				Reference soil, ppm in dry matter			
Treatment	Fe	Mn	Zn	Cu	Fe	Mn	Zn	Cu
DTPA A - 250	235	380	18000	15.0	148	238	104	12.5
B - 500	158	535	17500	5.0	126	216	84	10.0
Peat C - 5 %	580	835	14800	15.0	110	525	94	32.5
D - 10 %	128	772	12300	15.0	270	788	118	17.5
Lime E - 2000	168	238	2875	12.5	118	113	60	12.5
F - 4000	250	285	950	12.5	142	223	58	10.0
Control G	158	772	16300	5.0	110	185	82	10.0

SUMMARY AND CONCLUSIONS

Since the trace element composition of pasture crops is largely variable, several external factors influencing their absorption were studied :

- Application of varying levels of major element fertilizers under greenhouse conditions.
- Application of varying levels of trace elements under greenhouse conditions.
- Application of varying levels of Farm Yard Manure (F.Y.M.) and NPK fertilizers under greenhouse conditions.
- Long and short term fertilizer field experiments.
- Finally the problem of soil contamination was also considered, together with some factors influencing the mobility of contaminating elements.

1. Specific influence of varying levels of macro element on the trace element uptake by perennial ryegrass :

In spite of the buffering effect of the original quantities already present in the soil, different interactions between the major elements added and the trace elements in the plants were observed. The content and total uptake of trace elements by perennial ryegrass, as well as the yields, were remarkably influenced. The effect of the applied major element treatments started generally with the second cutting and this revealed to significant and highly significant regressions. Nitrogen and lime treatments had the most pronounced effect on the trace element situation of the plant. Particularly the elements B, Zn and Mn were more affected than Fe and Cu. The effect of K on B and Pb was remarkable.

2. Effect of varying levels of trace element fertilization on trace element uptake by grass grown under greenhouse conditions.

Significant responses of Mn, Zn, Cu, B, Co and Mo concentration by perennial ryegrass and total uptake in relation to trace

element applications, were recorded.
The pH of the soil was generally found to be a dominant factor with regard to the availability. Other factors were also reported.
Toxicity of trace elements was clearly observed in plants grown on sandy soils, and not on the ones grown on a sandy loam soil. The toxicity phenomenon went together with highly increased contents of most of the trace elements in the plant tissues. This increase however was somewhat leveled off in the soil with the highest pH, while also certain antagonisms were involved.
Interactions between Fe and Mn, as well as Fe and Ni ; could be confirmed through the study given in this chapter.

3. Influence of Farm Yard Manure (F.Y.M.) and NPK fertilization, applied in a sandy soil at different ratios, on the trace element uptake of perennial ryegrass under greenhouse conditions.

Highly significant regressions were observed for the elements Mn and Zn and, to a certain extend, for B. For Fe and Cu the concentrations in the plant tissues were generally not significantly influenced. The present results indicate that a considerable increase in dry matter production is not necessarily linked with lower mineral element contents and that the so called "dilution effect" is not to be generalised.
F.Y.M. treatments suppressed the concentration as well as the total uptake of Mn and Zn.
The trace element ratio between roots and shoots were also studied.

4. Effect of micro and macronutrient fertilizers on pasture crops (field experiments).

a. Long term experiments :

Long term one sided fertilization with major elements had an indirect effect on the trace element status of plants, partly due to changes in soil pH. The most sensitive elements are Mn, responding to slight pH differences, as well as copper, boron and lead. Toxic levels of copper was observed in the first harvesting, when a

mixture of trace elements (20 kg/ha) was applied as spray.
Also relatively high values of the other trace elements during the same cutting were recorded. The treatments giving the lowest dry matter productions systematically showed that highest trace element concentrations.
Application of trace elements for meeting the requirement of the plants is suggested to be based on the observation of mineral contents, soil characteristics, plant species. Seasonal fluctuations in trace element contents were also observed. This concerned the elements Fe, Mn, Zn and Cu. Their values positively correlated with the higher amount of rainfall recorded in 1968.
The antagonistic effect between Fe and Mn acted as an important parameter with respect to the absorption of these elements. Also their ratio appeared to be a largely influenced factor.
Particular attention should be drawn towards the contents of the separate species. The present results indicate that clover contained higher values of Fe, B and Pb than grass, while grass was higher in Mn and no systematic differences were observed for Zn and Cu.

b. Short term experiment

The short term field experiment showed less variation in soil pH, the values were more stable. Significant and highly significant concentration values for Fe, Mn, Zn, Cu, B, Pb and Ni were recorded when factors such as magnesium, nitrogen, phosphorus and potassium were studied.

5. Prelimenary observations concerning metal contamination in Belgian grassland.

The effect of mineral contamination on the soil-plant relationships under field and greenhouse conditions was studied. Mineral contamination of the soil influenced the growth as well as the mineral composition and quality of the plants. Perennial ryegrass (R.v.P.) was found to be more tolerant to highly contaminated soil with Pb, Zn and Cu than agrostis tenuis. In all cases, raising the pH of the soil by liming, appeared to be the most efficient factor for reducing the absorption of toxic trace elements. However the mobility of Fe and Mn was more pH dependent than that of zinc, copper and boron.

In the case of very high contamination levels, the usual liming rates, bringing the soil to neutrality, seem still to be insufficient for reducing the trace element uptake to normal values.

Contamination of Pb in soils and plants along the highways was clearly observed but remains localised within a limited distance. In cases of more generalised contamination however, the changes in crop composition are to be carefully observed in order to prevent hazardous effect with respect to animal nutrition.

Summarising, the numerous observations concerning the trace elements behaviour and their absorption by pasture crops, appears to be subject to a complex system of equilibria and interactions. Some of these permit to make quite reliable predictions, but in several cases, it seems that only experimental observations, based on analytical information, is actually the only way to judge about locally existing situations.

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